MYCOPATHOLOGIA

ET

MYCOLOGIA APPLICATA

FUNDATORES:
R. CIFERRI
P. REDAELLI

EDENDA CURAVIT:
TIBOR BENEDEK
Chicago, Ill.

EDITORES:

F. de Almeida, São Paulo — A. N. Aravysky, Leningrad — A. E. de Arêa Leão, Rio de Janeiro—E. Baldacci, Milano — J. Bánhegyi, Budapest — E. Baudys, Brno— A. L. van Beverwijk, Baarn — K. B. Boedijn, The Hague — S. R. Bose, Calcutta — Wm. Bridge Cooke, Cincinnati, Ohio — H. Campins, Barquisimeto, Venezuela — A. L. Carrión, San Juan, P. R. — A. Castellani, Lisboa — A. Catanei, Alger — K. Cejp, Praha — A. Chaves Batista, Pernambuco — A. Ciccarone, Bari — N. F. Conant, Durham, N. C. — J. N. Couch, Chapel Hill, N. C. — J. Coudert, Lyon — S. N. Dasgupta, Lucknow — E. D. Delamater, Philadelphia — C. W. Dodge, St. Louis, Mo. — C. W. Emmons, Washington, D.C. — E. Fejér, Budapest — G. W. Fischer, Pullman, Wash. — G. Florenzano, Firenze — A. da Fonseca, Rio de Janeiro — N. Fries, Uppsala — E. Gäumann, Zürich — A. Gonzalez Ochoa, Mexico, D. F. — H. Götz, München — L. Grigoraki, Marseille — E. I. Grin, Sarajevo — E. Hazen, Albany N.Y.— R. Heim, Paris — T. Hemmi, Kyoto — S. Ito, Sapporo — D. Janke, Marburg/Lahn— J. Jørstad, Oslo — P. N. Kashkin, Leningrad — F. D. Kern, State College, Pa. — A. M. Kligman, Philadelphia, Pa. — C. da Silva Lacaz, São Paulo — J. Lodder, Delft — J.E. Mackinnon, Montevideo — O. de Magalhaes, Belo Horizonte — A. v. Mallinkrodt-Haupt, Köln — P. Martens, Louvain — D. S. Martin, Durham, N. C. — J. Meyer, Strasbourg — M. Pereiro Miguens, Santiago de Compostela — J. H. Miller, Athens Ga. — R. Morquer, Toulouse — E. M. Mrak, Berkeley, Calif. — J. A. Nannfeldt, Upsala — P. Negroni, Buenos Aires — F. L. Niño, Buenos Aires — J. Obrtel, Praha — T. Ørskov, København — Pereira Filho, Porto Alegre — B. Peyronel, Torino — A. Pijper, Pretoria — A. Pilat, Praha — P. Pinetti, Cagliari — A. Pinto Viegas, Campinas — J. Politis, Athènes — G. Pollacci, Pavia — V. Puntoni, Roma — T. Rayss, Jerusalem — I. Reichert, Rehovot — F. Reiss, New York — E. Rivalier, Paris - D. P. Rogers, Urbana, Ill. — K. Sakaguchi, Tokyo — R. Sartory, Strasbourg — G. H. Scherr, Chicago — J. Schwarz, Cincinnati — F. J. Seaver, Winter Park, Fla. — G. Ségretain, Paris — R. Singer, Tucumán — A. H. Smith, Ann Arbor, Mich. — F. K. Sparrow, Ann Arbor, Mich. — J. A. Stevenson, Beltsville, Md. — Y. Takahashi, Sendai — R. V. Talice, Montevideo — M. J. Thirumalachar, Pimpri, Poona — A. Trotter, Vittorio Veneto — N. van Uden, Lisboa — A. C. Ukil, Calcutta — R. Vanbreuseghem, Anvers — C. Vermeil, Tunis — O. Verona, Pisa — G. A. de Vries, Baarn — S. A. Waksman, New Brunswick, N. J. — J. Walker, London — L. E. Wehmeyer, Ann. Arbor, Mich. — J. Westerdijk, Baarn.

VOLUMEN XII

UITGEVERIJ Dr. W. JUNK — DEN HAAG — 1960

VOLUMEN XII

Fasc. 1. (pag. 1— 96) edit: 30—XII—1959
Fasc. 2. pag. 97—176) edit: 25—II—1960
Fasc. 3. (pag. 177—256) edit: 31—III—1960
Fasc. 4. (pag. 257—368) edit: 25—V—1960
Supplementum edit: 15—IV—1960

INDEX AUTORUM

Ajello, L., 101, 199.
Alteras, I., 223.
Andleigh, H. S., 159, 278.
Benedek, T., 96, 170, 171, 172, 175, 177, 255, 360, 366, 368.
Beretta, G., 189.
Boedijn, K. B., 163.
Borzani, W., 97.
Briceño-Maaz, T., 199.
Bridge Cooke, W., 1, 168, 173, 363, 367.
Campins, H., 199.
Caretta, G., 233.
Chaves Batista, A., 185, 196.
Chmel, L., 77.
Ciferri, R., 251.
Crosby, P F., 145, 349.
Evolceanu, R., 216, 223.
Evron-Maoz, R., 111.
Farr, M. L., 283.
Fitzgerald, J. D. L., 257.
Goss, L. J., 101.
Götz, H., 124.
Gresham, G. A., 207.
Ito, K., 67.
Kaplan, W., 101.
Kuehn, H. H., 145, 349.
Landi S., 257.
Lažar, M., 216.
Margalith, P., 189.
Meyer, E., 254.
Moore, J. C., 199.
Muftic, K. M., 265.
Nino, F. L., 46.
Paldrok, H., 289.
Pessôa Coêlho, R., 185.
Raubitschek, F., 111.
Rêgo Vieira, J., 185.
Scherr, G. H., 364.
Silveira, J. S., 196.
Sue ivens, M., 101.
Tilak, S. T., 141.
Viswanathan, T. S., 156
Wehmeyer, L. E., 169.
Whittle, C. H., 207.

Copyright 1960 by Uitgeverij Dr. W. JUNK, Den Haag
Printed in Holland by
Dijkstra's Drukkerij n.v., voorheen Boekdrukkerij Gebroeders Hoitsema, Groningen

CONTENTS: CONTENUTO: INHALT: TABLE DES MATIÈRES:

Pag.

Wm. Bridge Cooke: An Ecological Life History of *Aureobasidium pullulans* (De Bary) Arnaud. *(Robert A. Taft Sanit. Eng. Center, Publ. Health Serv., U.S. Dept. of Health, Educ. & Welf., Cincinnati, Ohio, U.S.A.)* 1

F. L. Niño: A proposito del llamado micetoma maduromicosico del pulmon. *(Lab. del Inst. de Clín. Quirurg., Hosp. de Clín., Buenos Aires, Argentina)* 46

K. Ito: Experimental Fungus Granuloma as Seen from Comparative Exanthematology. *(Dept. of Dermatol., Gifu Med. Coll., Gifu-City, Japan)* 67

L. Chmel: Critical Survey of Medical Mycology in Czechoslovakia for the years 1946 to 1956. *(Derm.-venerol. Clin., Bratislava, C.S.R.)* 77

W. Borzani: Production of Crystals by *Rozites gonglyophora*. (*Dept. of Chem., Esc. Politecn., Univ. of São Paulo, Brasil*) 97

W. Kaplan, L. J. Goss, L. Ajello & M. Sue Ivens: Pulmonary Mucormycosis in a Harp Seal Caused by *Mucor pusillus*. (*Communicable Dis. Centre, Publ. Health Serv., Dept. of Health, Educ., Welf., Atlanta, Ga., USA*) . . . 101

R. Evron-Maoz & F. Raubitschek: The Morphology of the Pseudoparasitic Phase of Some Dermatophytes. (*Mycol. Lab., Dept. of Dermat. and Vener., Hadassah Univ. Hosp., Jerusalem, Israel*) 111

H. Götz: Zur Morphologie der Pilzelemente im Stratum corneum bei Tinea (Epidermophytia) pedis, manus et inguinalis. (*Derm. Klin. und Poliklin., Univ. München, Deutschland*) 124

S. T. Tilak: Two New Species of Pleospora From India. (*M.A.C.S. Lab., Poona, India*). 141

H. H. Kuehn & P. F. Crosby: Nutritional Studies of *Eidamella deflexa*. I. Carbon-Nitrogen Assimilation and Some Growth Factor Requirements. (*Grain Processing Corp., Muscatine, Iowa, USA*) 145

T. S. Viswanathan: A New Species of Mycosphaerella From India. (*M.A.C.S. Lab., Poona, India*) 156

H. S. Andleigh: Use of Achromycin in the Preparation of Selective Media for Fungi. (*S.M.S. Med. Coll., Jaipur, India*) 159

K. B. Boedijn: On a New Genus of the Endomycetaceae 163

IV. Venezuelan Congress of Phthisiology and Pneumology. Deep Mycoses in Venezuela with Special Reference to Pulmonary Mycoses, by **Tibor Benedek** 177

A. Chaves Batista, R. Pessôa Coêlho & J. Rêgo Vieira: Da ocorrência de *Hanseniaspora valbyensis* Kloecker em apêndice cecal humano e lesões epidérmicas. (*Inst. de Micol., Univ. do Recife, Brasil*). 185

P. Margalith & G. Beretta: A New Antibiotic Producing Streptomyces: *Str. bellus* nov. sp. (*Res. Labs. Lepetit, Milano, Italy*). 189

A. Chaves Batista & Jarbas S. Silveira: *Trichosporon figueirae* n. sp. assinalado como fungo intestinal. (*Inst. de Micol., Univ. do Recife, Brasil*) 196

Libero Ajello, Tulio Briceño-Maaz, Humberto Campins & Jane C. Moore: Isolation of *Histoplasma capsulatum* from an oil bird (*Steatornis caripensis*) Cave in Venezuela 199

C. Howard Whittle & G. Austin Gresham: *Candida* in vitro and in vivo 207

R. Evolceanu & M. Lazar: Le problème de l'existence et de la mise en évidence des ferments kératinolytiques chez les dermatophytes. (*Sect. Derm., Inst. de Thérapeut., Bucarest, Roumanie*). 216

R. Evolceanu & I. Alteras: De l'origine saprophytique, tellurique des dermatophytes: une espèce du genre *Aleurisma*, saprophyte du sol et agent d'une dermatomycose expérimentale. (*Sect. Dermatol., Inst. de Thérapeut., Bucarest, Roumanie*) 223

G. Caretta: *Coremiella cuboidea* (Sacc. et Ell.) Cif. et Car. n. comb. isolata da feci umane (*Centro di Micol. Med., Ist. Bot. dell'Univ., Pavia, Italia*) 233

R. Ciferri: About the "Precis de Mycologie" by the late M. Langeron (*Centro di Micol. Med., Univ. di Pavia, Italia*) 251

S. Landi & J. D. L. Fitzgerald: Sensitization and Anaphylactic Shock Induced in Guinea Pigs by Using Fungus Extracts. *(Dept. of Bot., Univ. of Toronto and Dept. of Allergy, Western Hosp., Toronto, Ont., Canada)* 257

Mahmoud Kamal Muftic: The Genus Blastomycoides as a Mycological Entity. (*Gaza, Palestine, U.A.R.*) 265

H. S. Andleigh: Investigations Into the Role of Fungi in Pulmonary Diseases. (*Reader in Path. and Bacteriol., S. M. S. Med. Coll., Jaipur, India*) . 278

Marie L. Farr: Mycological Notes. New Species, Combinations and Host Records. (*Crops Res. Div., Agr. Res. Serv., U.S. Dept. of Agric., Beltsville, Md., USA*) 283

Heiti Paldrok: Medical Mycology in Scandinavia and Finland During a Century, From 1841 To 1940. (*Dept. of Derm., Karolinska Sjukhuset, Stockholm, Sweden*) 289

H. H. Kuehn & P. F. Crosby: Growth and Nutrition of a Strain of *Gymnoascus reessii* in Shaken Cultures. (*Grain Processing Co., Muscatine, Iowa, USA*) 349

Libri Novi 96, 168, 254, 360

Gallery of Contemporary Noted Mycologists Nrs. 61 to 76.

Supplementum: O. Verona & T. Benedik: Iconographia Mycologica II: Plates A19—A46, B11—B23, C 19—C25, D5—D6.

GALLERY OF CONTEMPORARY NOTED MYCOLOGISTS

61. E. Rivalier, Paris.
62. Emil Mrak, Davis.
63. K. Cejp, Praha.
64. P. Pinetti, Cagliari.
65. I. Jørstad, Oslo.
66. C. J. La Touche, Leeds.
67. T. Dominik, Szczecin.
68. R. Singer, Tucemán.
69. Frank D. Kern, Philadelphia.
70. E. Fejér, Budapest.
71. Alfred S. Sussman, Ann Arbor.
72. Hernan Miranda, Trujillo.
73. E. S. Beneke, East Lansing.
74. Marvin A. Rich, New York.
75. R. Sartory, Strasbourg.
76. A. M. Kligman, Philadelphia.

PLATES: TAVOLE: PLANCHES: TAFELN:

I-II. P. Margalith & G. Beretta 192/193
III. K. M. Muftic 272/273
Gallery Nrs. 61, 62, 63, 64 . . . . 96/ 97
Gallery Nrs. 65, 66, 67, 68 . . . . 176/177
Gallery Nrs. 69, 70, 71, 72 . . . . 256/257
Gallery Nrs. 73, 74, 75, 76 368

131 text figures

AN ECOLOGICAL LIFE HISTORY OF AUREOBASIDIUM PULLULANS (DE BARY) ARNAUD

by

WM. BRIDGE COOKE

Robert A. Taft Sanitary Engineering Center
Bureau of State Services
Public Health Service
U. S. Department of Health, Education, and Welfare
Cincinnati, Ohio

(21.IV.1959)

INTRODUCTION

The fungus *Aureobasidium pullulans* (DE BARY) ARNAUD has been known for 90 years under a wide variety of names from a wide variety of habitats. It has been considered a plant pathogen, a human pathogen, a saprobe, a mold of nectar, of butter, of margarine, of buds, of decaying litter, and of many other habitats. It is commonly known as the "black yeast".

Among the students of soil populations, of the microbes of decay, and of laboratory contaminants, this fungus has been commonly called *Pullularia pullulans* (DE BARY) BERKHOUT. However, its importance to the student of the grape in France and Italy antedates other interests, and to phytopathologists in these countries it is known as *Aureobasidium pullulans*, although it has more nuisance than damage value. The editor of the Experiment Station Record (1894) admitted to knowing of the name *Aureobasidium*, but believed it was superfluous since, although he probably never saw this fungus, he believed it was really an *Exobasidium*.

The following report is a review of published information concerning this fungus. It is part of an attempt to determine the correct name and some of the bases of variation for the fungus as well as its position in nature. It is based on Outline III of "Ecological life history outlines for fungi" (COOKE, 1951). A rearrangement of the topics suggested in the outline seems appropriate for our approach here.

TAXONOMY

The species *Dematium pullulans* was described by DE BARY in

1866. The genus *Aureobasidium* was established in 1891 by Viala & Boyer for a fungus which appeared to be the cause of a scald of grape leaves. Both de Bary's species and Viala & Boyer's genus have been redescribed a number of times under various names for strains which appeared to be causing diseases of legumes and flax, and for molds appearing in various places including the honey of flowers. The genera in which it has been described are listed below and following them are listed the species and combinations proposed for different strains of this fungus found in diverse habitats. It will be noted that the fungus is ubiquitous and omnivorous. More intensive cultural studies of some of the strains found on plant tissues may demonstrate their independence from the saprobic strains and the strains reported to be pathogenic for man. Some of this synonymy is based on Ciferri, et al., 1956.

Aureobasidium Viala & Boyer, C. R. Acad. Sci. Paris **112**: 1148. 1891.
Sarcinomyces Lindner, Mikrosk. Betriebsk. Ed. **2**: 222. 1898.
Kabatiella Bubak, Hedwigia **46**: 297. 1907.
Protocoronospora Atk. & Edg., J. Mycol. **13**: 185. 1907.
Foxia Cast., Trop. Med. Hyg. **11**: 260. 1908.
Pachybasidiella Bub. & Syd., Ann. Mycol. **13**: 9. 1915.
Polyspora Lafferty, Proc. Dublin Soc. **21**: 258. 1921.
Monotropomyces Constantin & Dufour, C. R. Acad. Sci. Paris **173**: 957. 1921.
Exobasidiopsis Karak., Not. Syst. Inst. Crypt. Petr. **1**: 83. 1922.
Pullularia Berkhout, Schimm. Monilia, etc., Univ. Utrecht 55. 1923.
Rhabdogloeopsis Petrak, Ann. Mycol. **23**: 52. 1925.
Aureobasis Clements & Shear, The Genera of Fungi, 343. 1931.
Nomen nudum - Corrected spelling for *Aureobasidium.*
Protocoronis Clements & Shear, The Genera of Fungi, 344. 1931.
Nomen nudum Corrected spelling for *Protocoronospora.*

Aureobasidium pullulans (de Bary) Arnaud, Ann. Mycol. **8**: 475. 1910.
Torula oleae Cast., Cat. Pl. Env. Marseill. 222. 1845.
Dematium pullulans de Bary, Morph. Physiol. d. Pilze, Flechten u. Myxomyzeten 182. 1866.
Tubercularia ampelophila Sacc., Mich. **2**: 262. 1878.
Hormiscium oleae (Cast.) Sacc., Syll. Fung. **4**: 265. 1886.
Tuberculina? ampelophila (Sacc.) Sacc., Syll. Fung. **4**: 656. 1886
Aureobasidium vitis Viala & Boyer, C. R. Acad. Sci. Paris **112**: 148. 1891.
Exobasidium vitis (Viala & Boyer) Prilleuix & Delacroix, C. R. Acad. Sci. Paris. **119**: 106. 1894.
Aureobasidium australiensis McAlpine in Viala, Rev. Vitic. **5**: 588. 1896.
Aureobasidium vitis var. *tuberculatum* McAlpine, Addit. Fungi Vine, Australia. 16. 1898.
Aureobasidium vitis var. *album* Montemartini, Atti Ist. Bot. R. Univ. Pavia. II, 5: 71. 1898.
Oidium pullulans (de Bary) Lindner, Wschr. f. Brauerei **15**: 209—213, 1898.
Monilia variabilis Lindner, Mikrosk. Betriebsk., Ed. **2**: 222. 1898.
Sarcinomyces crustaceus Lindner, Mikrosk. Betriebsk. Ed. **2**: 222. 1898.
?*Sarcinomyces crustaceus* f. *album* Lindner, Mikrosk. Betriebsk. Ed. **3**: 301—302. 1901.
Sarcinomyces albus Lindner, Mikrosk. Betriebsk. Ed. **3**: 301—302. 1901.
?*Cladosporium* (*Dematium*) *chodati* Nechitsch, Inst. Bot. Univ. Genève, Ser. 6, fasc. 5: 22. 1904.
Hormiscium gelatinosum Hedgc., Mo. Bot. Gard. Ann. Rept. **17**: 101. 1906.
Oospora pullulans (de Bary) Sacc., Syll. Fung. **18**: 499. 1906.

Torula granulosa LINDAU in RABENH., Krypt. Fl. **1**(8): 579. 1907.
Oospora variabilis (LINDNER) LINDAU in RABENH., Krypt. Fl. **1**(8): 34. 1907.
?*Oidium moniliaeforme* WIEGMAN & WOLF, Cbl. Bakteriol. II. **22**: 659. 1909.
Monilia nigra BURRI & STAUB., Landw. Jb. Schweiz 487. 1909.
?*Monotoyella nigra* CASTELLANI & CHALMERS, Man. Trop. Med. Ed. **2**: 799. 1913.
Coniothecium crustaceum (LINDNER) NEGER, Flora **110**: 120. 1918.
Monilia fusca BROWNE, J. Ind. eng. Chem. **10**: 178. 1919.
Polyspora lini LAFFERTY, Sci. Proc. Roy. Dublin Soc. **16** N.S.: 258. 1921.
Monotropomyces nigrescens CONSTANTIN & DUFOUR, C. R. Acad. Sci. Paris. **173**: 957—959. 1921.
Trichosporium mantegazzae POLL., Riv. Biol. **4**: 313—328. 1922.
Pullularia pullulans (DE BARY) BERKHOUT, Schimmelgeschl. Monilia, Oidium, Oospora on Torula. 55. 1923.
?*Pullularia pullulans* var. *fusca* (BROWNE) BERKH., Schimmelgeschl. 55—56. 1923.
Pullularia sarcinomyces (BEIJERINCK) BERKH., Schimmelgesch. 55—56. 1923.
Pullularia nigra (BEIJERINCK) BERKH., Schimmelgesch. 55—56. 1923.
Cladosporium metanigrum (CASTELLANI) FERRARI, Atti Ist. Bor. R. Univ. Pavia IV. **3**: 183. 1932.
Hormonema dermatoides LAGERBERG & MELIN, Skogsof. Tidskr., Stockholm. **25**: 219. 1927.
Cryptococcus metaniger CASTELLANI, Arch. Dermat. Syph. **16**: 402. 1927.
Hormonema pullulans (DE BARY) LAGERBERG & MELIN in ROBAK, Nyt. Mag. Naturwidensk. **71**: 256. 1932.
?*Torula lecanii-corni*? BENEDEK & SPECHT, Zbl. Bakt. I. **130**: 86. 1933.
Hormiscium dermatitidis KANO, Arch. Dermat. Syph. **176**: 282. 1937.
Torula schoenii ROUKHELMAN, C. R. Soc. Biol. Paris **124**: 547. 1937.
?*Pullularia crustacea* (LINDNER) DELITSCH, Syst. Schimmelpl. 123. 1943.
Pullularia botryotrichum DELITSCH, Syst. Schimmelpl. 123. 1943.
Pullularia granulosa (LINDAU) DELITSCH, Syst. Schimmelpl. 124. 1943.
Pullularia pullulans var. *lini* (LAFFERTY) WHITE, J. Aust. Counc. Sci. Ind. Res. **18**: 149. 1945.
Fonsecaea dermatitidis (KANO) CARRION, Arch. Dermat. Syph. **61**: 1007. 1950.
Hormodendrum dermatitidis (KANO) CONANT, in CONANT et al., Ed. **2**: 276. 1954.
Pullularia fermentans WYNNE & GOTT, J. gen. Microbiol. **14**: 516. 1956.
Pullularia fermentans var. *fermentans*, J. gen. Microbiol. **14**: 516. 1956.
Pullularia fermentans var. *melinii* WYNNE & GOTT, J. gen. Microbiol. **14**: 517. 1956
Pullularia fermentans var. *leaoi* WYNNE & GOTT, J. gen. Microbiol. **14**: 517. 1956.
Pullularia fermentans var. *schoenii* (ROUKHELMAN) WYNNE & GOTT., J. gen. Microbiol. **14**: 517. 1956.
Pullularia fermentans var. *benedekii* WYNNE & GOTT, J. gen. Microbiol. **14**: 518. 1956.

Pullularia werneckii (HORTA PARR.) DE VRIES, Contrib. Knowl. gen. Cladosporium. 101. 1952.
Microsporum mansoni CAST., Brit. med. J. **2**: 1271. 1905.
Foxia mansoni (CAST.) CAST., J. trop. Med. Hyg. **11**: 260. 1908.
Cladosporium mansoni (CAST.) PINOY, Ann. Derm. Syphil. V. **3**: 341. 1912.
Cladosporium werneckii HORTA PARR., Rev. Med.-Chirurg. Brasil **22**: 269. 1921.
Torula mansoni (CAST.) VUILLEMIN, C. R. Acad. Sci. Paris. **169**: 406. 1929.
Melassezia mansoni (CAST.) VERDUN (1912) in NANNIZZI, Rep. Sist. 405. 1934.
Dematium werneckii (HORTA PARR.) DODGE, Med. Mycol. 676.1935.
Dematium mansoni (CAST.) DODGE, Med. Mycol. 678. 1935.
Torula werneckii sensu COUDERT, Guide Prat. Mycologie, Med., Paris. 321.1955.
Pullularia fermentans var. *castellani* WYNNE & GOTT, J. gen. Microbiol. **14**: 518. 1956.

Two specific taxa appear in the literature as *nomina nuda* since they were apparently never published but only appear on cultures at Baarn according to BERKHOUT. These are: *Dematium nigrum* and *D. sarcinomyces.* Apparently both were laboratory names used by BEIJERINCK. REDAELLI & CIFERRI (1947) published an annotated account of human pathogenic fungi in which the incorrect spelling *Aerobasidium pullulans* was used. If this taxon is considered to have been published, it, too, is a *nomen nudum.*

Although CIFERRI (1956) included the *Cladosporium mansoni-werneckii* complex as conspecific with *A. pullulans,* it is being kept separate for reasons to be considered later.

DESCRIPTION

Colonies white at first, remaining white or usually becoming black throughout except at the margin, or with blackened areas developing irregularly, in radiating lines or sectors, as marginal areas or as concentric rings in all or part of the colony; colonies composed of a mat of mycelium appearing mucoid in many cases but also yeasty or dry, dull or shiny; hyphae radiating from the center of the colony or the point of inoculation, becoming irregularly intertangled, branched irregularly but usually at right angles, formed of elongate to spherical cells; young hyphae and hyphae in young slide cultures with elongate cells 1.5—2.5 × 20 μ or more long, becoming broader and shorter in older portions, 10—20 × 4—6 μ, and in many areas replaced by elongate chains of spherical to broad ovate cells reaching 10—15 or even as much as 48 μ in diameter in certain media; young hyphae thin-walled, hyaline, hyphae in maturing areas becoming thick-walled; septa in young hyphae simple, in older hyphae becoming thickened and double; cells and hyphal segments in older hyphae breaking apart at the thick, double-walled septa to form arthrospores or arthrospore-like hyphal segments; thick-walled ovate or spherical cells appearing like chlamydospores or papulospora-like aggregations of cells; all cells capable of producing conidia on papillae produced from the inner wall of the cell at irregular intervals; occasional papillae becoming short hyphal branches resembling false basidia on which irregular numbers of papillae produce conidiospores; conidiospores hyaline, ovate, drawn out at the base to a point where attached to the papilla, 1.5—2 × 4—5 μ in slide cultures on young mycelium, reaching 6 × 12 μ in older areas of the mycelium, averaging 3—4 × 7—8 μ; conidiospores giving rise to secondary conidia by yeast-like budding, yeast-like spores averaging slightly smaller than primary conidiospores; all hyphae at first hyaline, walls becoming pale green to olive green to brown and finally black in age in irregular areas or throughout most of the cells of a mycelium; throughout a black colony, or black portions of other colonies, occasional cells remain thin-walled and hyaline; conidiospores mucoid. Description based on a culture (SEC 3105) on neopeptone-dextrose agar.

The above morphological description is subject to considerable variation from strain to strain, from culture to culture, from medium to medium, and even from one batch of synthetic medium to another. In part, this helps to explain the large number of names applied to this fungus. This variation indicates either an extremely unstable type of fungus or a highly heterocaryotic condition in which many possible strain types are represented in a single culture.

Endogenous conidia (KLOCKER & SCHOEINNING, 1900) are produced under certain conditions as a result of proliferative growth within the walls of dead cells from living cells at one, the other, or both living ends. Rich culture media and high humidities are required. This is not considered to be a normal type of growth.

CLASSIFICATION

CLEMENTS & SHEAR (1931) were unfamiliar with this fungus. They placed *Pullularia* in the heterogenous assemblage "Pseudosaccharomycetes" while the genus *Aureobasidium* was included in the Hypochnaceae (Basidiomycetes) with *Exobasidium* and, apparently as a special precaution, in the Melanconiaceae of the Fungi Imperfecti. BOUTHILET (1951) found that "black yeasts" were difficult to identify because of inadequate taxonomic treatments.

If one is using the Saccardoan system as outlined by CLEMENTS & SHEAR, a more rational approach would be to include *Aureobasidium* in the Fungi Imperfecti, Order Moniliales, Family Dematiaceae, Subfamily Amerosporae. For those who prefer the system developed by VUILLEMIN, LANGERON & TALLICE (1932) and later NEGRONI & FISCHER (1942) suggest that this fungus should be classified in the Fungi Imperfecti, Hyphomycetes, Order Thallosporales, Suborder Blastoarthrosporineae, Family Trichosporaceae; or as CASTELLANI & DE SILVA (1957) suggest: Thallosporales, Blastosporineae, Cladosporiaceae. However, LANGERON & GUERRA (1938) include it in their discussion of medical yeasts only as a mold. According to the writer's interpretation of his system, this genus would fall in section IB of HUGHES' interpretation (1953) of the Hyphomycetes. The conidia are considered to be blastospores (slimy radula spores according to MASON, 1937), and on this basis the fungus was placed in the Amerosporae of the Gloeosporae by WAKEFIELD & BISBY (1941). DIDDENS & LODDER (1942) place this fungus in that section of the Dematiaceae with yeast-like cells. On occasion the mycelium apparently can break up into arthrospores or arthrospore-like segments. This fragmentation is not as general, nor as complete, throughout the mycelium as that found in *Geotrichum*, and may be related more to the chlamydospore nature of the thick-walled mycelial cells than to their arthrosporic characteristics. MRAK & PHAFF (1948) state that

"— when identifying yeasts it is difficult to exclude such commonly encountered genera as *Pullularia*, *Geotrichum* or *Sporobolomyces* —".

ARNAUD (1910) was the first writer after VIALA & BOYER (1891) to use the generic name *Aureobasidium* and to define its characteristics. It was not until 1956 that CIFERRI reintroduced *Aureobasidium* to the literature, although it had been used occasionally in listing or describing specimens. In 1951, WHITE wrote to ROGERS concerning the validity of the genus and wondered whether *Aureobasidium* was available for use. ROGERS (1951) replied that he was unfamiliar with it as a Basidiomycete.

It is suggested that the reason for the confusion over the name of this organism is based in part on a certain amount of artistic license practiced by the early illustrators of the genus. Apparently to prove that the spore bearing structures they saw could be called basidia, using a loose definition of this structure, VIALA & BOYER (1891) drew only 4 spores attached to the hyphal projections they thought of as basidia. PRILLIEUX (1895), in illustrating this fungus which he had transferred to *Exobasidium* the previous year, showed erect spore bearing cells with usually 4 spores around their tips. A few basidium-like conidiophores which were illustrated had spores produced on the sides as many strains of *Aureobasidium pullulans* do. MCALPINE (1898) definitely illustrated 4 and 6 spored basidia in his description of *A. vitis* var. *tuberculatum*. However, from his description and discussion it is evident that he was studying *A. pullulans*. While *Hormonema* was the first generic name published with a Latin diagnosis among the synonyms of *Aureobasidium*, and thus considered the correct name by the Swedish workers (ROBAK, 1932), the rule requiring a Latin diagnosis did not become effective until 1935 so that the earliest published generic name in the list of synonyms presented above would apply correctly.

NATURAL APPEARANCE AND PHENOLOGY

In general, this fungus is inconspicuous in its natural habitat. It cannot be readily identified in rough mounts of mycelium and fruiting materials from many types of habitat, since it has cells which may be readily confused with those of other species. In connection with contamination of painted surfaces, and in wood, the identification of this fungus *in situ* may be rendered less difficult since it is frequently the only organism present, or its structure is distinctive. In grape leaves, on flax plants and in the nectar of flowers, it may be less difficult to identify because it may be the only fungus present. In these as well as in other habitats it must be isolated and studied in pure culture when its biological relationship with other members of a population can only be the subject of conjecture.

Because the fungus is ubiquitous and, to a large extent, omnivorous, its phenology can be stated rather simply; following spore germination on a favorable habitat, mycelial production is initiated, conidia are produced if the nutrient supply is adequate, and growth and conidial production continue until the nutrient supply available to it is exhausted. Following exhaustion of nutrients, or treatment of substrata with inhibiting substances (see below), conidial production is halted and either a resting stage is initiated or death occurs. The conidia may be air-borne and at least in some areas (MORROW, 1943) may occur in large enough numbers to be considered as "showers" after observation of exposed plates. This rough outline of a life history is given as a phenology. For most strains, dates cannot be supplied since growth may be initiated at any time suitable substrata are available to spores. In case of grape and flax diseases, or growth on nectar, dates are more rigid and the fungus is dependent on the condition of the host tissues.

HABITAT

The numbers and types of places from which *Aureobasidium pullulans* has been isolated are large and varied. According to records, this fungus has become adapted to such a variety of habitats that one cannot be surprised at the large number of names it has received during the last 92 years.

In Australia (Anon., 1941; JENSEN, 1941, 1944) and Holland (WIERINGA, 1956) it has been found to be a common organism associated with the dew-retting of flax, a process through which flax fibers for linen manufacture are separated from other stem tissues by microbiological activity. JENSEN studied dew-retting under laboratory conditions and found two distinct cultural types of *Dematium pullulans*. Both types were active and activities were best at 30° C. Good retting was produced after 12—14 days. There was a definite succession of fungi on flax straw during the retting process but during active retting *D. pullulans* was the active organism. Other molds are potentially active in the retting process but *D. pullulans* appeared to be capable of suppressing them. The same group of molds is dominant in retting as is associated in the sooty mold community on leaves.

In Washington State, ENGLISH (1940, 1945) and ENGLISH & GERHARDT (1943) have found that this fungus, among others, was the fourth most important mold involved in the decay in storage and transit of pears, and third commonest isolate from lesions on cherries. While *Pullularia* was not found on immature cherries from Lewiston, Idaho, and Hood River, Oregon (ENGLISH, 1945), it was noted that it could be expected on mature fruit from those areas. It was commonest on fruits from Wenatchee, Washington, and 87 % of the damage it caused was associated with rain injuries. It was weakly pathogenic, produced no aerial mycelium, and did not spread by contact. TAYLOR & SHANOR (1945) reported it as a cause of a storage rot of tomatoes in West Virginia and Maryland. BENEKE, WHITE & FABIAN (1945) found that it was one of the more important fungi on strawberries in Michigan. *D. pullulans* was found by ADAM in 1923 to be one of 7 common fungi which produced molded oranges during the 3 months required for shipment from Australia to England. He found that resistance in oranges could be increased by "sweating" or allowing the orange rinds to dry out shortly before shipment. In Germany (MOLL, 1930) it was found that a source of mold inoculum in fruit sheds was mold growth in the pasteboard ceilings. *D. pullulans* and other molds spread to packing cases, fruit baskets, and other infectable types of equipment. RICHTER, in Germany (1931) found that when the moisture content of grain exceeded 15 % *D. pullulans* was one of 9 common fungi to produce mustiness and moldiness which made the grain unfit for feed or seed.

As a result of work with dew-retting of flax, SMIT & WIERINGA (1953) and WIERINGA (1953, 1955) were able to show that this mold is one of the first invaders of leaves which have fallen from trees and other plants, and that as a result of the activity of molds of this type the cutin of the leaf surface is broken down permitting the entry of other decay-producing organisms. This phenomenon may be related to factors recently pointed up by MARTIN, et al. (1957) in England, who showed that in the cutin of apple and other leaves are substances which are inhibitory to the growth of fungi during the life of the leaf. KEENER (1950) found in Pennsylvania that one of the commoner molds occurring on the buds of various types of woody plants was a species with the cultural characteristics of *Pullularia*.

On the other hand, certain fruits and leaves become subject to attack by this and other fungi even before their death or harvesting, or harbor it or its spores during the later stages of ripening. Whether this is because the fungus can counteract the inhibitory substances in the leaves, whether certain leaves or fruits do not possess such substances, or whether at maturity or on injury to tissues such substances become inoperative, has not been determined. Under at least two names this fungus has been reported as a weak pathogen of the leaves of grape in France and Italy (VIALA & BOYER, 1891; VIALA, 1893; PRILLIEUX & DELACROIX, 1894; GABATTO, 1907; and ARNAUD, 1931), Russia (FOEX & VIALA, 1896), Germany (SVOLBA, 1932), Poland (POTEBNIA, 1897), and Australia (MCALPINE, 1898). It has also been reported as causing a fruit decay in grape berries and in species of the genus *Prunus* (ARNAUD, 1931). In a recent monograph of the species previously assigned to the genus *Gloeosporium*, a cause of anthracnose of many kinds of plants, VON ARX (1957) separates species which produce spores in a manner similar to that of *Aureobasidium* into three genera: *Microstroma*, *Kabatiella* and *Aureobasidium*. Twenty species of saprobes and weak to mild plant pathogens are distributed among these genera.

A disease caused by this organism which results in the browning of stems of flax has been reported from as widespread locations as Ireland (LAFFERTY, 1921; MCKAY & LONGHAMS, 1947), and New Zealand (Anon., 1940; BAYLIS, 1941; and NEWHOOK, 1947), although the oil flax variety Rio appeared to be immune in New Zealand (Anon., 1940). In Poland (KONOPACKA, 1931), the fungus caused minor outbreaks associated with wet conditions. Experiments in Russia developed techniques of seed infection for studies involving varietal resistance. The method described by BAKUNOVA (1935) gave 99.3 % infection of mature plants. KAZINA (1935) found that if only 8.5 % of the seed were infected this did not appreciably affect the quality and yield of fiber and seed, but with 100 % infected seed there resulted a 20—25 % loss of fiber and 30 % loss of seed. The first symptoms of disease appeared at the bud formation stage when breaking of stems at the base was initiated. Since basal stem break was the point of entry of *Fusarium lini* the plants are rapidly killed. When PAVLOUSHIN (1937) tested for varietal resistance to *Polyspora lini* he found the reaction of the plant could best be observed in the period between the bud and flowering stages.

FLOR (1936a and b), in the United States, found that prior to 1932 the disease was known only in Michigan, but that by 1935 it had spread to Iowa, Minnesota, North Dakota and Oregon. Stem canker was severe in Iowa. Of the seed varieties Bison was most susceptible. Yield was reduced to 61 % of normal with seed weighing only 85 % of the controls. Tests showed (1936b) that seed treatment was not warranted in the northern Plains states.

WHITE in Tasmania (1945) considered *Polyspora lini* and *Pullularia pullulans* as cogeneric on the basis of comparative studies with six strains from Tasmania, New Zealand, Australia and Ireland. The fungus was considered weakly parasitic and non-pathogenic.

Working in Ireland, MUSKETT & MALONE (1941) developed a plate technique for determining presence of seed disease in a sample of seed. A sample of seed was mixed and seeds were removed with sterile forceps to malt extract agar in Petri dishes. After 5 days incubation at 22° C plates were examined for mold colonies around the seeds. In 1948, MUSKETT proposed a simple method in which transmitted light was used for detection of disease. COLHOUN & MUSKETT (1948) found that fungi of infected flax seed could live 15—55 months. The higher the percentage of infected seed, the longer it took the fungi to die. Prolonged seed storage was not recommended as a control measure.

Two fungi have been closely associated in rye-grass blind seed disease. It was not until 1940 that the differentiation of these two fungi was definitely established. In the meantime, HYDE (1938) in New Zealand reported that low germination rates of rye-grass seeds was due to infection of the kernel by *Pullularia*. If 80 % of the seed were infected the crop would be worthless as a seed source. Methods of laboratory tests for infected seeds were described. Infected seed heads have a sticky fluid containing conidiophores. If the seed is dry the colorless or pale pink secretion solidifies and has a waxy consistency. If rain or dew has washed the spores away the grain is dull rusty red. Examination of one area of rye-grass at 7—10 day intervals gave 10 %, 28 %, 44 % and 56 % infected seed with a germination rate of 37 %.

In England (NOBLE, 1939) it was found that infected seeds become opaque, that only 15—17 % of opaque seeds germinated in contrast with the very high germination rate of the translucent seeds. When seeds became very dark only 8 % germinated, although 40 % of the paler seeds germinated.

WILSON, et al. (1940a), in England, noted that the imperfect state of a *Helotium* which parasitized rye-grass seeds was similar to *Pullularia* and that the two fungi usually occurred together. MUSKETT & CALVERT (1940) showed that two fungi were definitely involved and that *Pullularia* was only a contaminating saprobe. WILSON, et al (1940b), repeated MUSKETT & CALVERT'S observations. WILSON et al. (1945), showed that the oily consistency of the slime of the conidial stage of *Phialea temulenta* distinguished this fungus from *Pullularia pullulans*.

In Australia (MASSEE, 1898) this fungus has been found associated with gummosis of *Prunus* spp. In California it was found associated with diamond canker of var. Standard of the French prune (Anon., 1930; SMITH, 1929). There is no indication that it was the cause of the canker. LEWIS (1912) studied this fungus in relation to its incidence in *Sphaeropsis malorum* leaf spots and cankers of apple but found its activity to be only saprobic.

Since the spores may occur on grapes, as well as other fruits, it is not surprising that CAPRIOLETTI (1956) has found this mold in grape must in Italy. LINDNER, as early as 1888, cautioned against the misinterpretation of the cells of *Dematium* in wort with or without hops for yeast cells. CARINO-CANINA (1929) sowed spores of molds in grape must and incubated them 50 days at 24° C. *Dematium pullulans* diminished the total acidity without producing an equivalent amount of volatile acids. It attacked dextrose and levulose.

Another type of leaf infection results from the colonizing activities of a group of fungi known as sooty molds. In many cases these colonies of fungi are associated with the aphid-type insects which inhabit the leaf rather than with the leaf surface itself. The surface of the leaf is probably used by the fungus as a place for anchorage rather than a source of nutrient. *Dematium, Aureobasidium,* or *Pullularia pullulans* has been reported as an important member of this population, sometimes one of the primary colonizers, in France (ARNAUD, 1910), Holland (TENGWALL, 1924), Germany (NEGER, 1918), and Australia (FRASER, 1933).

In discussing *Cladosporium lauri*, a new parasite of scale insects of the laurel, RAYBAUD (1923) indicated that *Dematium pullulans* was one of the associated molds but of little importance. This fungus was found to be the almost exclusive member of a sooty mold complex associated with the scale insects *Eulecanium corni* on the leaves of peach (*Prunus domestica*) in the Kamp Valley in Lower Austria. SVOLBA (1932) reported that the effects of these two organisms caused considerable damage in central Europe. The symbiotic relationship between this fungus and various plant pest insects is considered later.

HADDOW (1941) reported from Ontario that *Pullularia* followed gall midge injury in red pine (*Pinus resinosa*). The fungus penetrated the base of the leaf through insect punctures. Its effects are usually noted in forest nurseries rather than in natural vegetation in the National Forests. A strain of *Pullularia pullulans* isolated from lateral and terminal buds of *Pinus resinosa*, and which dissolved pith of living twigs of this species, could be recovered 5 weeks later as much as 1—2 cm from the injection sites (JUMP, 1938). The fungus was shown to exert a growth promoting effect on infected pines whose shoots grew more rapidly and produced abnormal bifurcations. In a 50-year old plantation of *Pinus strobus* in Nort Carolina (DOOLITTLE, 1948) *Pullularia pullulans* was found associated with a needle blight. More trees were infected on dry thinned sites than on moist unthinned sites. In England (BALKO, MURRAY & PEACE, 1938), *Sclerophoma pithyophila* was associated with a needle cast of pines. Evidence to indicate a connection between this species and *Pullularia pullulans* was obtained when surface sterilized leaves from healthy pines were inserted in cultures from infected needles. In about 90 % of these cases *S. pithyophila* developed on the needles. In Wisconsin, GRUENHAGEN, RIKER & RICHARDS (1947) found that this mold was involved in brown blight of red and jack pines after spittle insect injury. However, PATTON & RIKER (1954) decided on the basis of inoculations that *Pullularia pullulans* was associated only saprobically with this condition, that needle-blight was a gall-midge injury and needle-droop a physiological disorder. MOLNAR (1958a) has found in British Columbia that after aphid injury by *Pineus* sp. *Pullularia* was associated with a flagging of western white pine (*Pinus monticola*). In the latter case, proof of association was confirmed by isolation and reinfection experiments. *Pullularia pullulans* was present in 95 % of the white spruce trees showing necrosis following spruce budworm (*Choristeneura fumiferans* (CLEM.)) defoliation in the Babine Lake area of the Prince Rupert Forest District in British Columbia.

A substrate rich in carbohydrates and other nutrients is nectar of flowers. COSTANTIN & DUFOUR (1921) in France isolated this fungus from the flowers of *Monotropa* under the name of *Monotropmyces*. CIFERRI, et al (1956), in Italy found it in the flowers of weed and cultivated *Trifolium repens*, *T. incarnatum* and *Lilium tigrinum*. However, *Aureobasidium pullulans* was associated with flowers and other parts of inflorescences of date palms in Libya which had rotted away from the trees as a result of activity by a *Fusarium* (CIFERRI, 1939).

Slime fluxes of trees are rich in sugars and other nutrients and attract a special population. Studies have been made occasionally on one or another member of this population, but in England (OGILVIE, 1924) a study has been made of the populations. It was found that *Dematium pullulans* was a common member of the mold populations of the brown variety of the Brown Slime Flux which was usually found on elm and horse-chestnut trees. It was not isolated from other varieties of the brown, or from other types of slime fluxes.

From the different habitats listed above and their wide geographic separation, it is obvious that this is a widely distributed organism. This is further borne out by air sampling experiments. In sampling air throughout the world, the spores of this organism are among those found more commonly. MORROW (1947) and her co-workers (1942, 1943) have indicated something of this in their reports of air surveys and summaries of published work on the air "spora". The occurrence of *Pullularia* spores in air is limited in some areas and at some seasons, but there is no pattern of conformity for it is more often found as "showers" of spores than as a regular member of the population. Apparently these showers are not correlated with season or climate and no effort has been made to determine the source of the spore showers developed from plate or slide exposures. To date it has not been demonstrated that the fungus is important in allergy. Other areas in which the fungus has been reported as a member of the air spora are Sweden (RENNERFELDT, 1947), Minnesota (SWEABLY & CHRISTENSEN, 1951), New Zealand (DYE & VERNON, 1952), and England (MAUNSELL, 1954). Except in spore showers, percentage occurrence is usually small but the presence of the organism in samplings is consistent. It may become abundant enough so that it represents between the third and tenth commonest fungus found. In sampling high altitude air (9000 ft.) over Canada, the Arctic and the Atlantic Ocean, PADY and his co-workers (1951—1957) have found this fungus wherever exposures have been made with plates or slides. The fungus is not readily identifiable from visual observation of spores deposited on slides but must be observed among developing colonies on agar in Petri dishes. It is thus obvious that spores trapped at high altitudes and high latitudes are viable, whether they were produced locally or were brought into the region in masses of air from more southern latitudes.

Spores settling from the air may have traveled long distances as the work of PADY has shown. Such spores, settling on substrata which contain nutrients for growth of the fungus, initiate colonies and develop as scavengers in the soil, on wood or other material, or they may develop as nuisance organisms in places where their activity is not favorable to the physical or economic life of man.

Among scavengers, this fungus plays a small role in the soil as a member of the large population of soil microorganisms. Its numbers are low but *Pullularia* or *Dematium pullulans* has been isolated from many soil samples tested for fungi. It has been reported from the United States by WAKSMAN (1917), MARTIN (1956), GILMAN (1957), HERR (1957), and MILLER, et al. (1957); from England by DALE (1914), CHESTERS (1948), and JEFFREYS (1953); from Germany by NIETHAMMER (1941); from Egypt by SABET (1935), and from Australia by WARCUP (1955, 1957). BOUTHILET (1948) studied 84 samples of Minnesota soils for yeasts. These were collected during one spring season. He obtained 142 cultures of yeasts of which 25 were of this black yeast. It is usually found in soil particles as mycelial fragments. From the soil it may be introduced in run-off water to sewage treatment plants from which it has been reported by BECKER & SHAW from Washington and Idaho (1955), and by COOKE from Ohio (1957).

Pullularia pullulans has been isolated, among other yeast-like fungi and yeasts, from the surfaces of shrimp in the Gulf of Mexico (PHAFF, MRAK & WILLIAMS, 1952), indicating that at least some strains are salt water tolerant.

From the fact that its spores occur in air and that it occurs in soil, it should not be surprising that it is found as a contaminant of wood being processed for lumber and for paper pulp. As one of the organisms causing blue stain of saw timber and of pulp wood, it has been reported from Sweden where much of the work on this phase of its existence has been done (LAGERBERG, et al. 1927; MELIN & NANNFELDT, 1935; BJÖRKMANN, 1956). In some of this work it has also been reported from Finland. LAGERBERG, et al. (1927), report it as extremely common, occurring at the same time on living leaves and other parts of the living host plant, and that it is important in the blue stain of water floated logs. BJÖRLING (1937) found at least 5 bluing fungi in at least 11 localities in Sweden growing in symbiosis with algae on tree bark. Mycelial elements were intertwined with 1, 2 or 4 algal cells with haustoria appearing only on the surface of algal cells. This was considered a potential source of infection for timber and pulp wood. It is an exception to the rule that it is impossible to draw up a safe working definition based on its morphology in wood. HEDGCOCK (1906) reported it in the United States under the name *Hormiscium gelatinosum.* VERRALL (1939) found it infrequently on pine and not at all on hardwoods in mill ponds in Louisiana, Mississippi and Georgia. CIFERRI, et al. (1956), found it on old wood in Italy.

MALM (1941) reported *Pullularia pullulans* as occurring in paper mill slimes in Sweden. From its behavior in culture in the laboratory at Cincinnati it is easy to understand how it would become troublesome in this habitat. In shaker culture and in stationary liquid culture, in most media, it forms tightly adhesive mats of slimy mycelium on the sides of glassware and is difficult to separate from its nutrient supply through membrane or asbestos filters under 25 pounds of vacuum. RENNERFELT (1946) found that pinosylvin and pinosylvin monomethyl ether from the heartwood at concentrations of 0.02 % inhibited the growth of this fungus in malt agar. It was thus more apt to produce bluing in sapwood than in heartwood.

HUBERT (1929) emphasized the importance of blue stain fungi in wood used in various types of interior and exterior finishing such as doors, window sash, etc. He presented keys to different species and methods of control. *Hormiscium gelatinosum* was one of the species considered. DAVIDSON (1935) considered this fungus of little importance on new wood in the southern states but of more importance on old wood. HUBERT (1938) summarized work on blue stain fungi and listed *Pullularia pullulans* and *Hormiscium gelatinosum* as two of the causative fungi. These were resistant to control by the methods in use at that time. Using *H. gelatinosum* as the test organism, HUBERT (1940) described methods for evaluation of oil-soluble wood preservatives. Minimum requirements for toxicity of the fungus were given. Using tests based on *H. gelatinosum* MELLECKER & BAKER (1942) found that painted wood showed no significant fungal growth after dipping treatments. SHEFER (1946) listed general fungicides for blue stain fungi using chemical dipping treatments for stain fungi on boxing and crating wood. In flask cultures with Scots pine sapwood blocks, BJÖRKMAN (1947) found that blue stain fungi gave some protection against wood rot fungi but that these fungi do not prevent attack of wood by rot producing fungi. Blue stain fungi appear to be more tolerant of millwork preservatives than wood rot fungi according to experiments by KRAUSE (1948). Using a modification of the National Door Manufacturers' Association tests, it was found that 0.5 % phenylmercuric oleate and a combination of this with 0.1 % zinc naphthenate containing 1 % metallic zinc were most effective against blue stain. That dryness is needed in the control of stain and rot fungi was shown by SCHAEFER & CHIDESTER (1948). In wood with a moisture content of 12 % stored in a conditioning room at 65 % relative humidity and 80° F. *H. gelatinosum* survived for less than a year. FISCHER (1948) indicated that pentachlorophenol was toxic to stain and rot fungi at concentrations of 20 p.p.m. The material was incorporated in finishing emulsions.

ROBAK (1932) has presented extensive studies based on this fungus which he called *Hormonema pullulans*. He described 8 strains and indicated that in his experience (Scandinavian) this was one of the most active wood pulp staining fungi, its activity being demonstrated only a few days after inoculation. SCHMID (1935) found this fungus to be one of 6 species involved in staining.

Control possibilities were limited by economic considerations. In Finland, BADE & KLEM (1935) related various types of pulp grinding equipment in Scandinavian paper mills to the problem of fungal infections. *D. pullulans* was one of the fungi considered. Isolations on glucose or malt agars (RENNERFELT, 1937) indicated that *P. pullulans* was one of the commonest blue stain molds in pulp. It could be found commonly in the wood itself, fresh water, air, and, most important back water where logs were stored. Newly manufactured pulp from a closed system was more subject to infection than that from an open system. Frozen pulp was more susceptible after thawing. Bluing fungi were more resistant to high and to low temperatures. Only *P. pullulans* and *Trichosporium heteromorphum* (*Margarinomyces heteromorphum*) were able to multiply by budding during the first 48 hours of incubation at 1°, 20°, and 35° C, but chiefly at the medium temperature. Sanitary controls were considered. Green and floated wood should not be mixed in pulp ponds according to RENNERFELT (1939a) who found that the growth of *P. pullulans* was more rapid on green than floated wood, and better on extracts of green wood than on glucose agar. ENGLAND (1939) thought that biological control of the common *P. pullulans* had more prospects for future success than chemical control. He found that this fungus grew best out of doors at 20°—30° C, but in the plant it grew well at 35°—65° C. During processing, wood pulp is infected in such a way (RENNERFELT, 1939b) that 10—40 % of the spores in the white water are filtered with the fibres on the kamyr machine. The large number of spores in the white water do not go to the pulp but return to the system. *P. pullulans* is an aerobe, so if the air is driven out of a bale of pulp (AIROLA, 1941) damage from aerobic organisms is potentially reduced.

SÉE (1917, 1919) in France has found *Dematium pullulans* to be a mold which attacks the paper of old books. It has also been found in the Archives of Poland (KOWALIK, 1952) to be an important mold causing the deterioration of paper.

In work with deterioration of various types of materials *Aureobasidium pullulans* has been picked up in association with other fungi from many different kinds of supplies. In Germany, HANSAM & LIEBSCHER (1940) found it causing spots on sheep leather treated with synthetic tanning preparations. RICHARDS (1949) in New York found that this was one of the common molds contaminating the surfaces of optical instruments. Direct inoculation was unsuccessful, but if natural contaminants such as this are left on the glass they produced etchings and raised places. Cleanliness in manufacture and throughout the life of the instrument, keeping the surface free from nutritious substances, storage at above room temperature in cabinets to lessen humidity, and application of fungicides, are methods of prevention of mold contamination.

Wherever materials of any kind subject to mold action are exposed without protection, mold will grow on them. Some of these molds are actively degrading the materials, others live on degradation products or impurities in the materials. In testing various types of materials accidentally exposed to the elements, discarded, in use but not protected, or deliberately exposed to microbial action, *Pullularia pullulans* has appeared commonly. It grew (Anon., 1945; GRAY, 1945; WESTON, 1949) on such items as canvas webbing, trousers, leather scabbards, cotton duck, gloves, tent ropes, shoe leather, canteen bags, belts, raincoats, and tentage found or exposed in such widely spaced areas as New Guinea, Guadalcanal, Panama Canal Zone, and Florida. It was also reported on a tyroglyphid mite in a binocular from the Canal Zone. BARGHOORN (1945) found *P. pullulans* to be one of the numerically more important fungi in 23 samples exposed in sunny locations in Panama.

To determine whether or not such fungus contaminants are causing damage to the materials on which they have been found, test procedures (WHITE, et al. 1948) have been developed. early testing was reported by GALLOWAY (1930) who found no deterioration of cellulose filter paper in mineral salts solution from strains isolated from two types of cotton yarns. CUNNINGHAM (1940) used Shirlan WS to control mildewing of tentage and tarpaulins in which *P. pullulans* was mildly involved. Degradation tests with strains picked up in Florida and New Guinea (WHITE, 1945) proved negative. GRAY (1945), WHITE, et al. (1948), and MARSH, et al. (1949), found that *Pullularia* strains which were tested gave little if any activity on cellulosic materials indicating that in nature they were living on the products of metabolism of other organisms or in impurities in the cloth or leather goods.

In tests for fungicides, *P. pullulans* was used (MELORO, 1950) to check the antifungal activities of fire resistance- and copper-treated cotton duck. Fire-resistance material successfully resisted fungal attack while copper containing compounds produced different degrees of inhibition.

Occasionally this fungus becomes a contaminant of foods. HERTER & FORMET (1919) found it in bread in France; WIEGMANN & WOLF reported it from butter in Germany (1909), and CIFERRI, et al. (1956), isolated it from butter and margarine in Italy. It has been found in Germany (MAURIZIO & STAUB, 1928) as a cause of black specking of Emmenthaler cheese. BROOKS & HANFORD (1923) reported that it was one of the molds which contaminated cold-store meats in England. SKINNER, EMMONS & TSUCHIYA (1947) report fungi similar to this species causing a black mold of raw sugar and black spotting of yeast cake. ALWOOD (1908) isolated this species from a laboratory contaminated sugar solution while studying the fermenting power of yeasts and associated fungi. A solution of "mycoban", calcium propionate, was prepared for use in laboratory work at Pullman, Washington. On examination of a contaminant identified as *P. pullulans*, which developed during the first month's storage at room conditions it was found that this fungus was able to develop in the solution. This substance is used in the manufacture of certain foods such as bread to prevent rapid development of molds.

Njoku-Obi, et al. (1957), found that after chickens were treated with Chlortetracycline and placed in refrigerator storage they tended to spoil. Among the fungi isolated from treated, but not from untreated, chickens was *P. pullulans* (*Torula nigra*). Chickens were dressed at the State College of Washington poultry processing plant with a mechanical scalder and picking machine. After scalding at 140° F (60° C) for 40 seconds they were warm-eviscerated, halved, cooled in ice water for 2 hours, and then immersed in a solution with 20 p.p.m. chlortetracycline hydrochloric acid for 10 min., drained, placed on paper board trays, enclosed in plyethylene bags and stored in a mechanical refrigerator at 34—36° F (1.1—2.2° C). After 6 days daily observations were made. Presence of slime and spoilage odors indicated spoilage. Dipping 500 gm. spoiled chicken in 500 ml water gave a reporting ratio of 1 : 1 for microorganisms found. After dipping, carcasses were discarded and the wash water diluted to 1/10, 1/100 or 1/1000. One ml of each dilution was plated in peptone-beef extract-glucose-tartaric acid agar and incubated at 25° C for 5 days.

Food packaging materials are subject to fungus activity and through this portal fungi can get to the food. This is especially true of butter. In New Zealand (Riddett & Mill, 1936) butter is packed in wood boxes for shipment to England. *P. pullulans* was found to be one of 5 common molds which grow on the surface of such containers and thence onto the butter. Its growth was stimulated by defrosting the frozen packaged butter under conditions of high atmospheric temperature and humitity. Growth was better when air could reach the inside of the box, and when the wood was rotary cut instead of sawn. Wood used in such boxing, locally called "white pine", *Podocarpus dacrydioides*, was not readily susceptible to fungus attack but became increasingly so upon exposure to these conditions. Swedish pine wood impregnated with wax was less susceptible but gave a peculiar flavor to the butter. Aluminum foil placed between two pieces of parchment when used as a wrap prevented molds from reaching the butter.

In New York, Sandborn (1937) reported that food packaging papers could be contaminated by various molds from the slimes in paper mills during the manufacturing process. Certain chemical methods could be used to control slimes in early stages of production but immediate casing or sealing of finished paper, the avoidance of direct human contact with such paper, and normal hygienic procedures should be used in later production stages.

Dematium pullulans has been reported to be one of the common molds of wine cellars where high humidities and careless handling provide adequate environmental conditions for its growth.

Painted and varnished surfaces and coatings are subject to infestation by *P. pullulans*. The fungus produces black patches in white paint similar to those obtained from excessive fumes of H_2S. HAENSLER (1921) in New Jersey, and REYNOLDS (1950) in Florida, have described activities of this type, and REYNOLDS has published an extensive discussion of such injuries, the causal organism, and how to prevent their recurrence, VICKLUND & MANOWITZ (1951) found that this fungus is the commonest species on painted surfaces where it grows on the oils in the paint. In Germany, MEIER & SCHMIDT (1952) found that they could isolate a black *Dematium* from painted surfaces by removing scrapings to 14 day old linoxylin, incubating for 4 weeks, and transferring to other media for further growth.

It has been found (GOLL & COFFEY, 1948) that *P. pullulans* is the most common mildew of interior and exterior paints. It becomes deeply embedded in the paint film and thus disrupts the paint continuity. It is important in surface coatings in breweries, distilleries, textile mills, and other types of factories. It is difficult to accelerate its speed of growth artificially as well as to replicate out of doors conditions in the laboratory.

A mildew test of interior paints has been developed by the Nuodex laboratories (1955). Untreated or treated paint is applied on two successive days to 6 × 10″ filter paper. The paper is then leached in water for 24 hours at 80—85° F and air dried. One and a quarter inch squares are cut, dipped in boiling water for 30 seconds, dipped in a spore suspension, incubated 28 days at 26—28° C and 80—85 % relative humidity. Six different fungi are recommended for this test including *P. pullulans*. The growth on the specimen is recorded in 12 categories of coverage and density. Exterior paint is tested in the same way except that only *P. pullulans* is used as a test organism.

A different type of fungus activity attributed to this species may be mentioned. Several pathogenic conditions of man, among which Tinea nigra palmaris is the most prominent, have been attributed to this fungus. Cutaneous fungus infections in which this species has been implicated have been reported from Colombia (MONTOYA & FLOREY, 1898), Argentina (NEGRONI, 1942), Puerto Rico (CIFERRI & ASHFORD, 1929; CARRION, 1950), and Tripoli and Italy (REDAELLI & CIFERRI, 1947). In addition, CASTELLANI & CHALMERS (1913) have listed a number of different types of disorders from many parts of the world in which this fungus was considered to be either the causal organism or a contaminant of more than usual importance. Recently, WYNNE & GOTT (1956) in Texas obtained *Pullularia* from lymph nodes in a case of Hodgkin granuloma. It is of interest to note that the fungus has not proved to be pathogenic in animal tests with strains isolated from human disease cases.

Castellani & de Silva (1957) have presented an interesting review of the history, distribution, taxonomy, experimental reproduction of disease, symptomatology, diagnosis, prognosis and treatment of disease produced by a *Pullularia*-like fungus which they assign to *Cladosporium*, but which de Vries (1952) considered a species of *Pullularia.* Apparently a person does not acquire immunity from the disease since de Silva was able to inoculate himself successfully on three different occasions. The incubation period was found to be 15 days. The fungus grows in such layers of the skin as the stratum corneum and the stratum granulosum. Diagnosis is relatively easy since the fungus can be cultured readily from biopsy material. The general health of the patient is not affected, but in the tropics there is no tendency toward a spontaneous cure. However, persons in the tropics who contract the disease produced by this fungus in the skin apparently lose it on moving to temperate regions.

Since 1884, when Metchnikoff described a yeast parasite of a species of the crustacean *Daphnia*, yeasts and yeast-like fungi have been known to be associated with animals, especially insects. At first, these organisms were considered parasitic and Moniez (1887) described *Lecaniascus polymorphus* as causing a disease in the scale insect *Lecanium hesperidium.* Lindner (1895) added to this list of apparently parasitic yeasts by describing *Saccharomyces apiculatus* var. *parasiticus* from *Aspidotus nerii.* Berlèse (1906) was the last of this group to consider these organisms parasitic when he described *Oospora saccardiana* from *Ceroplastes rusci.*

The first mycoentomologist to think of those fungi which occur in various parts of the insect body as symbionts was Šulc (1906) who described a new genus and two new species of yeast-like fungi from two coccoid scale insects. Most of the insects carrying such symbionts are sucking insects such as scales, aphids and cicadas. Buchner (1912) organized some of the information about them. Šulc (1910) and Buchner (1912) added more yeast-like species and additional host insects to the growing list so that by 1912, 18 species of such fungi had been described in 7 genera. Breest (1914) discussed the problem of fungus-insect symbiosis, listing some of the insects known to possess such a relationship, but apparently considering the nameless fungi as variants of a single species. Teodoro (1918) added another species. In 1921, Buchner summarized the information he had accumulated in one of the chapters in his book, revised in 1953, on intracellular symbiosis in plants and animals.

Brues & Glaser (1921) placed a fungus they found in the fatbodies of the scale insect *Pulvinaria innumerabilis* in the genus *Dematium* after finding it did not agree with species which had been described earlier. They indicated that Moniez's fungus (mentioned above) was probably the organs described by Leydig in 1854 as "pseudonavicellae".

BRAIN (1923) brought the systematic side of this picture to a climax in a paper in which he described 41 species of these fungi in 11 genera. The generic groupings appear to be based on groups of hosts. There were no cultural studies and species within genera appear to be based upon more or less minor variations in morphology within the host group. The illustrations indicate an ability of the fungi to become adapted to the environment of the particular host as much as demonstrating specific variation.

A review (Anon., 1925) of a paper by PIERANTONI (1925) notes that the suggested term for organs in which symbiotic organisms tend to congregate is mycetome. STEINHAUS (1947) noted that ŠULC (1910) coined this term and that he referred to single cells carrying such organisms as mycetocytes. Such organs or organ-like aggregations of tissue or cytoplasm are found in secretory glands, intestinal walls, and, in the case of luminous organs, in the skin.

A special group of symbionts has received some attention in the case of lacquer-producing scale insects. In *Lakshadia indica* MADIHASSAN, a coccoid scale insect associated with "stocklak" production, TSCHIRCH (1925) described a symbiotic fungus, *Laccomyces symbioticus*, which was thought to be related to the lac-producing ability of the insect. Apparently this was a renaming of the organism described by LINDNER in 1895. In 1928, MADIHASSAN described in general terms the symbionts of and waxes produced by wax and pseudo-lac insects in India. No names were used and, except for size, no special variations were noted. No culture studies were made by this worker either in 1928 or in 1929 when he reported on red and yellow lac insects. Symbionts appear to belong to *Bacillus* and *Nocardia* among the bacteria and to *Torula rosaceae* in the fungi. These organisms are not considered to be related to the lac-producing abilities of their hosts by MADIHASSAN. Symbionts of 6 scale insects are illustrated but were considered to be members of the same species.

BENEDEK & SPECHT (1933) found a fungus, *Torula lecanii-corni*, as a symbiont of the scale *Lecanium corni* in Germany. They used hanging drop cultures to isolate the fungus from the insect and the strains they obtained agreed in all respects with the fungus in the original hanging drop cultures. CIFERRI, et al. (1956), used the available strain of this fungus in comparative studies of 23 strains obtained from culture collections and their own isolations and decided it was a strain of *Aureobasidium pullulans*.

STEINHAUS (1947) was the first to bring together in English all the information on fungus-insect symbiosis available to him at that time. His survey is a very thorough one.

In 1952, STEINHAUS noted that yeast-like symbionts occurred with three scale insects in the San Francisco Bay area. In 1955 he reported on studies made on these and on an additional insect symbiote in which an attempt was made to discover the physiological requirements of the symbiotes so that this information could be used in upsetting the symbiotic balance between the two organisms, thus effecting a type of biological control of the scale insects which usually cause considerable damage. The symbiotes were not obtained in pure culture since efforts in this direction "met with no certain success beyond the limited growth and budding of individual symbiotes held in hanging drop preparations". *Pullularia pullulans* was a common associate of the host insects being studied and, in spite of all precautions, was commonly cultured from all the insects studied. This fact, and the fact that "in many respects the symbiotes of the coccoids studied are suggestive of the so-called black yeasts of which *Pullularia pullulans* is the best known species", led Steinhaus to make the following suggestion: "It is not impossible that the symbiotes in the insect have a phylogenetic relationship to the free-living *Pullularia*, and that long ago the symbiotic arrangement grew out of the constant association between the insect and the *Pullularia* in its environment."

At this point it would be appropriate to introduce the names of the genera and species to which various insect symbiotes have been assigned. This list is not added to the main list of synonyms *Aureobasidium pullulans* because relationship is only suspected, not proved, and cannot be proved without pure culture studies. The order of listing is strictly chronological and no genus or species should be considered as preferred over another in the list.

Genera:

Lecaniascus Moniez, Soc. zool. France Bul. **12:** 150—152. 1887.
Saccharomyces sensu Lindner, Zbl. Bakt. II. **1:** 782—787. 1895.
Kermincola Sulc, Sitzber. Kgl. Böhm. Ges. Wiss., Art. **19:** 1—6. 1906.
Oospora sensu Berlèse, Redia **3:** 8—15. 1906.
Cicadomyces Šulc, S. B. Kgl. Böhm. Ges. Wiss., 1910 **(14):** 1—6. 1910.
Coccidomyces Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Aleurodomyces Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Schizosaccharomyces sensu Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Psyllidomyces Buchner, Arch. Protistenk. **26:** 1—116. 1912.
Lecaniocola Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Physokermincola Brain, Ann. Univ. Stellenbosch **1(A)(11):** 25. 1923.
Cicadocola Brain, Ann. Univ. Stellenbosch **1(A)(11):** 25. 1923.
Cissococcomyces Brain, Ann. Univ. Stellenbosch **1(A)(11):** 27. 1923.
Iceryomyces Brain, Ann. Univ. Stellenbosch **1(A)(11):** 28. 1923.
Aphidomyces Brain, Ann. Univ. Stellenbosch **1(A)(11):** 28. 1923.
Chermomyces Brain, Ann. Univ. Stellenbosch **1(A)(11):** 30. 1923.
Laccomyces Tschirch, Handbuch der Pharmakognosie, Leipzig **3(2):** 965. 1925.

Species:

Lecaniascus polymorphus Moniez, Soc. zool. France Bul. **12:** 150—152. 1887.
Saccharomyces apiculatus var. *parasiticus* Lindner, Zbl. Bakt. II. **1:** 782—87. 1895.
Oospora saccardiana Berlèse, Redia **3:** 8—15. 1906.
Kermincola kermesiana Šulc S. B. Kgl. Böhm. Ges. Wiss., Art. **19:** 1—6. 1906.
Kermincola physokermincola Šulc, S. B. Kgl. Böhm. Ges. Wiss., Art. **19:** 1—6. 1906.
Saccharomyces macropsidis-lanionis Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—29. 1910.
Saccharomyces conomeli-limbati Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—29. 1910.

Saccharomyces cicadarum Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(14):** 2. 1910.
Cicadomyces ptyeli-lineati Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—29. 1910.
Cicadomyces ptyeli-lineati form II Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—29. 1910.
Cicadomyces aphalarae-calthae Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—29. 1910.
Cicadomyces cicadarum Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 5. 1910.
Cicadomyces aphrophorae-alni Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—39. 1910.
Cicadomyces aphrophorae-salicis Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—39. 1910.
Saccharomyces aphalaeae-calthae Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—39. 1910.
Saccharomyces aphidia Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—39, 1910.
Schizosaccharomyces psyllae-foesteri Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—39. 1910.
Schizosaccharomyces chermetis-strobilobii Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—39. 1910.
Schizosaccharomyces chermetis-abietis Šulc, S. B. Kgl. Böhm. Ges. Wiss. **1910(3):** 1—39. 1910.
Cicadomyces sulcii Buchner, S. B. Ges. Morph. Physiol. (München) **27:** 89—96. 1911 (1912).
Saccharomyces anobii Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Coccidomyces rosae Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Psillidomyces tenuis Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Cicadomyces minor Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Cicadomyces liberiae Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Cicadomyces minimus Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Cicadomyces dubius Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Cicadomyces rubricinctus Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Coccidomyces dactylopii Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Coccidomyces pierantonii Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Schizosaccharomyces drepanosiphi Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Schizosaccharomyces sulcii Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Aleurodomyces signoretii Buchner, Arch. Parasitenk. **26:** 1—116. 1912.
Lecaniocola parasitica (Lindner) Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola putnami Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola contei Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola rosae (Buchner) Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola filippiae Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola inglisiae Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola pulvinariae Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola protopulvinariae Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola proteae Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola lecanii-viridi Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola egbarum-fulleri Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola saccardiana (Berlèse) Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola cercoplastidis-pallidi Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola (?) *macropsidis-lanionis* (Šulc) Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Lecaniocola (?) *conomeli-limbati* (Šulc) Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Kermincola tenuis (Buchner) Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Physokermincola physokermina (Šulc) Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Cicadocola cicadarum (Šulc) Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
Cissococoomyces natalensis Brain, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.

eryomyces pierantoni (BUCHNER) BRAIN, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
phidomyces aphalarae-calthae (ŠULC) BRAIN, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
phidomyces aphidis (ŠULC) BRAIN, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
phidomyces drepanosiphi (BUCHNER) BRAIN, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
phidomyces psyllae-foersteri (ŠULC) BRAIN, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
phidomyces sulcii (BUCHNER) BRAIN, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
hermomyces chermetis-strobilobii (ŠULC) BRAIN, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
hermomyces (as *Charmomyces*) *chermetis-abietis* (ŠULC) BRAIN, Ann. Univ. Stellenbosch **1(A)(11):** 21. 1923.
accomyces symbioticus TSCHIRCH, Handbuch der Pharmakognosie, Leipzig **3(2):** 965. 1925.
ecaniocola saissetiae GRANOVSKY, Wisc. Acad. Sci. Arts Lett., Trans. **34:** 445—56. 1929.

In summary, it may be said that BUCHNER (1953) terminated is discussion of studies on insect symbionts with the work of ENEDEK & SPECHT. The only symbiont mentioned by name was he earliest one described. In this recent edition of his text the iology of the symbiosis between yeast-like organisms and insects s considered more important than the taxonomy of the symbionts. t may be said that the latter aspect of the problem appears to ave been a fad without much permanent significance.

PHYSIOLOGY

Some studies have used *Aureobasidium pullulans* as a test rganism for observations on the ability of a fungus to use one or nother type of nutrients. Such studies have been based on the esire of a student for information about a specific fungus or a pecific substrate or type of substrate. The earliest study of this ype found was that by WAKSMAN (1916) who noted that this ungus could accumulate ammonia in the mycelium after 12 days rowth on ammonia-rich media. It could obtain half as much itrogen in growth on blood meal as on cottonseed meal. Old cultures were found to be only 1/3 to 2/3 as efficient in their nitrogen tilization as young cultures.

Nitrogen fixation by microorganisms is a problem which has nterested students for a long time. For more than 60 years yeasts nd pseudo-yeasts have been used in such investigations. LÖHNIS & ILLAI (1896) grew *Dematium pullulans* in a medium containing nannite. In the presence of $CaCO_3$ after 3 weeks a Kjeldahl deternination showed 0.41 mg N/100 cc of solution while without $CaCO_3$ .52 mg N/100 cc of solution was obtained. This work has been ited by some (LIPMAN, 1911-12; and GUILLERMOND & TANNER, 920) as evidence that the fungus can fix elemental nitrogen. n a more comprehensive study, BAUER (1938) found that this ungus was an oligonitrophile since in the absence of nitrogen in the medium some growth took place although it was considered that the fungus could not fix nitrogen. More recently (1942) SCHANDERL indicated that *Dematium pullulans*, together with 5 other molds, could assimilate atmospheric nitrogen. In an experiment with cultures of the fungus in a liquid mineral salts medium, using a Kjeldahl technique for determining nitrogen, there was an added 188.7 mg of nitrogen in the flask after 84 days, or an increase of 31 %. In a second experiment, at pH 6.0, there was an increase of 121.9 mg nitrogen or 25.7 %.

That *Pullularia* is assumed to be an obligate aerobe is indicated by the fact that only one reference has been found (LAGERBERG, et al., 1927) in which its oxygen tolerances are discussed. In relation to blue stain of wood, these workers found that while O_2 must be present, it can tolerate less O_2 and more H_2O than other fungi causing blue stain in water floated saw logs. Of the species tested, it required least O_2.

Dematium pullulans has been used in studies on pentose sugar utilization (ROSA, FRED & PETERSON, 1929). One strain of *Dematium* and one of *Monilia nigra* destroyed 66—98 % of the two pentoses used in the experiment. These were xylose and arabinose. Principal products of xylose were CO_2, alcohol and cellular material.

In a study on the fermentation products of *Dematium pullulans* carried out in Japan (SUMIKI, 1929), the following medium was used (the amounts given being in percentage units): glucose, 7.2; peptone, 0.1, or $(NH_4)_2SO_4$, 1.0; K_2HPO_4, 0.05; $MgSO_4$, 0.01; $CaCl_2$, 0.01; $FeCl_3$ T, and NaCl T. Cultures were incubated at 30° C for several weeks. The following products were identified: ethyl alcohol, acetic acid, succinic acid and *dl*-lactic acid. Quantities were small and most of the glucose was not consumed. The maximum yields of ethyl alcohol from 7.2 grams of glucose were 0.98 weight per cent, and 0.27 weight per cent.

ROUKHELMAN (1937) found that *Torula schoenii* could attack starch, sucrose, glycerol, glucose, lactose and maltose. The brown pigment in the older cells was found to be phenolic in nature and carbohydrates were stored in the cells as glycogen.

Important contaminant fungi of the milk industry were used in experiments on their physiology in relation to milk supplies in a study by CHRZASZCZ & SCHILLAK (1936). *Dematium pullulans* was found to be unimportant although it was frequently encountered. Best growth of this fungus was produced on wort agar after 26 days. After 21 days on calcium oxylate, 7.5 g mycelium were produced in ten cultures. By-products of growth included 5.95 % formic acid, 4.24 % acetic and propionic acids, no oxalic or citric acids, 15.7 % calcium carbonate, 13.41 % other products and mycelium. Traces of acetaldehyde and acetone were found. It was doubtful if fumaric and malic acids were produced, and no ethyl alcohol, glycol acid, glycoloxylactic acid, succinic or butyric acids were produced.

It has been reported (BIRKENSHAW, 1937, cited by WOLF &

WOLF, 1947) that *Dematium pullulans* mycelium contains as much as 0.3 % ergosterol.

In 1938, BAUER developed a careful study on the physiology of *Dematium pullulans* in which he observed the effects of certain nutrient combinations on both the morphology and the physiology of the fungus. Similar reactions were given by the five strains used. The mycelial phase was more pronounced under conditions of good aeration, a pH range of 3.66—3.91, 0.2—2.0 mol. $NaNO_3$ or 0.1—0.9 mol. NaSCN, and a bios-rich medium. Conidial production was more pronounced under poor aeration, pH 2.17—2.9 and a bios-poor nutrient. Conidial producing mycelium was formed in bios-poor synthetic media with such carbon sources as potato starch, soluble starch, mannit, glycerin, ethyl alcohol, fructose and dextrose, and nitrogen sources such as ammonium chloride and ammonium nitrate. In a strongly acid reacting medium, pH 3.0 and below, chlamydospore clusters of *Coniothecium*-like structure are formed in bios-poor media with the nitrogen sources listed above and the following carbon sources: dextrose, sucrose, fructose, maltose and ethyl alcohol. Abnormal growth forms were produced in high concentrations of nutrients: "hedgehog" colonies were produced in cultures with 2.21—2.54 mol. raw sugar, while spherical colonies were produced in 0.7—0.9 mol. NaSCN or 1.4—2.0 mol. $NaNO_3$. In the same concentrations of nitrogen compounds, but under strongly acid conditions (pH 1.92—3.00.) or strong alkaline conditions (pH about 10.1) abnormal giant cells were produced. At pH ranges of above 3.0, and in 0.1 mol. NaSCN or 0.2 mol. $NaNO_3$, slime production was reduced. At pH 3.0 or lower, abnormal melanin production occurred. Since the fungus can grow in conditions of high concentrations of sugars or of neutral salts, it appears to be well adapted to an existence in nature where such situations may exist naturally as in the nectar of flowers or the "honeydew" of the sooty-mold habitat. Its ability to grow as a filamentous fungus or as a yeast-like fungus under various conditions of aeration further increases the range of habitats in which it may occur. The wide pH range in which it can survive, pH 1.9—10.1, is another remarkable feature and may increase its adaptability to many environments.

SCHOPFER (1938) found that *Dematium nigrum*, among other fungi, was favorably acted upon by a homologue of thiamin in which the pyramidine fraction contained in position 2 an ethyl group in place of a methyl group, although the fungus was not deficient for thiamin itself.

RENNERFELT (1939—1940) studied the abilities of some blue molds of wood to grow on special media. *Pullularia pullulans* grew slowly and produced only hyaline cells on 1.5 % agar with 2.0 % pure glucose and Boas salt mixture. Better growth was obtained on this medium when technical grade glucose was used, and best growth was obtained on 1.5 % agar to which commercial malt extract was added. From the fact that variations were quantitative, it was assumed that malt extract may be richer in growth substances than technical grade glucose which in turn is richer than pure glucose.

In trying to determine if *Polyspora lini* and *Pullularia pullulans* were cogeneric, WHITE (1945) compared 6 cultures on various media. Two *Pullularia* cultures were obtained from Australia and Tasmania while four *Polyspora* cultures came from Australia, New Zealand and Ireland. No growth occurred on methyl red medium. On glucose, maltose, levulose and saccharose, growth was good and acid was produced by all strains. On *d*-xylose a strain of *Polyspora* from Victoria produced poor growth with no acid, while the other strains grew fair to well with acid production. On galactose, citric acid, succinic acid, tartaric acid, mannitol, glycerol and salicin no acid was produced and all strains grew but growth was variable. In an attempt to separate these strains by serology no definite pattern of serologic reaction was established on the basis of agglutination at a dilution of 1 : 450. Only host relationship would warrant maintaining strains assignable to *Polyspora lini* as a separate entity to be named *Pullularia pullulans* var. *lini*.

Penicillium glaucum and *Pullularia pullulans* were found growing in 21 days on a cocarboxylase solution which was 90 % hydrolysed to free thiamine (FARRER, 1949). These fungi were then grown on beer wort for 31 days at 20° C and phosphatase solutions were prepared by Seitz filtration. The *Penicillium* phosphatase was 62.5 times as active as that from *Pullularia pullulans*, 1 ml being as active as 600 mg of Parke-Davis Taka-diastase.

Pullularia pullulans has been found (WIERINGA, 1953) to be a predominant organism in the decomposition of pectin, thus preparing the way for other organisms of decay to enter leaves and twigs covered by cutin and other pectinaceous compounds. The apparent degradation of strawberries is associated with a greater pectinase content of affected berries. Pectolytic activity, as measured by per cent loss of viscosity, indicate (BENEKE, et al., 1954) that *Pullularia pullulans* can reach 50 % in 60 hours.

In studies with the antibiotics produced by microorganisms in acid heath soils, *P. pullulans* was found to be incapable of using carboxymethylcellulose (JEFFREYS, et al., 1953).

Nitrogen relations in well aerated media were investigated by LUTERAAN (1954a, b). When a balanced nitrogen supply (LUTERAAN 1954a) occurs in a well aerated medium, growth increases with a minimum consumption of glucose. Aeration may accentuate the effects of restriction of nitrogen supply: by slowing down growth, by lowering the percentage of nitrogen in the fungus, by raising the ratio of glucose consumed to nitrogen consumed faster than the ratio of glucose consumed to dry weight obtained, and by production of both fixed and diffusible melanin pigments. When insufficient oxygen supply limits growth, the ratios of glucose consumed to dry weight and percent nitrogen are raised.

Development of *P. pullulans* from the chlamydospore and the mycelial states varies according to the nitrogen supply. Growth in the presence of $NaNO_3$ is much slower and glucose consumption greater starting with chlamydospores than with a mycelium inoculum. The addition of small quantities of cysteine to the nitrate produces different development patterns for each inoculum type. Production of the two types of inoculum differs since a well aerated culture gives predominantly mycelial material while a poorly aerated liquid culture will give a chlamydospore inoculum. Cultures were grown in 100 ml medium in 1 L. Roux bottles. The medium consisted of essential mineral elements, nicotinamide as a source of growth factors, nitrogen source at N/18.75 and glucose at M/3.75. The cultures were carried for 7 days at 26° C. The initial pH of 3.9 reached 4.8 at the end of 7 days. Cysteine was added in some cases at a rate of M/300. In the case of mycelial inoculum, surface growth was obtained although cysteine retarded this. The chlamydospore inoculum gave a yeast-like culture with weak surface development. Weight yields were four times higher from the mycelial inoculum. When cysteine is present, yields from the chlamydospore inoculum increase, from the mycelial inoculum decrease, equalizing the differences in culture without cysteine. Other variations which occurred in these four cultures types related to rate of glucose utilization in the presence of different nitrogen sources although no fermentation products were formed.

Using a mineral salts basal medium, HENDERSON & FARMER (1955) studied the ability of *P. pullulans* to utilize several complex components of humus materials in the soil. *p*-Hydroxybenzaldehyde at an initial concentration of 0.01 % was completely removed. Vanillin was largely used up, less than 0.005 % remained of an initial concentration of 0.01 %. Of an initial concentration of 0.005 % ferulic acid, less than 0.001 % remained at the end of the experiment. In all cases the organism was exposed to the medium for 21 days at 21.5° C. Syringaldehyde was not used at all by this fungus.

Four strains of *P. pullulans* were cultured in coconut oil and ricinoleate (castor oil) (REESE, et al., 1955). On coconut oil, one strain produced between 11—100 mg of growth per flask or 3—20 % of the weight of oil used, two strains gave 101—200 mg growth or 21—40 % of the weight of oil used, and the fourth gave 201—300 mg growth or 41—60 % of the weight of oil used. In ricinoleate, two strains gave results of the lower type, while the other two gave results of the intermediate type.

In studying insect symbiotes, STEINHAUS (1955) found that *P. pullulans* was a very common fungus associated with scale insects and investigated some of its nutrient requirements and morphophysiological characteristics. The strains used assimilated glucose, sucrose, maltose, raffinose, and insulin, but did not assimilate lactose. Under proper conditions it fermented glucose to alcohol as determined by the Conway test for alcohol. Acid and gas were formed in glucose, sucrose, maltose and raffinose. Some strains assimilated ethyl alcohol slightly and some produced starch in varying amounts. Mycelium was formed after three days in glucose medium. Blastospores (buds) may be elongate and clustered, and the mycelium may branch at right angles. Blastospores may reach $4 \times 8\,\mu$ and sometimes are produced on protuberances. In sucrose these buds reach $2.5 \times 4.7\,\mu$ and may be borne singly or in clusters on protuberances. Yeast-like cells on malt agar vary with age: after 24 hours they average $4.5 \times 11.5\,\mu$, after 3 days $2.5 \times 10\,\mu$ and after 7 days $3.5 \times 15\,\mu$ Fourteen of 20 larvae of *Colias philodice eurytheme* BDVL. were killed by injection of spores but no deaths occurred following oral administration. Serological identification of internal symbionts with pure cultures failed.

"Fermentation" of sugars was used as a criterion for the varietal separation of strains of *P. fermentans* by WYNNE & GOTT (1956). Since previous descriptions did not include information about the fermentative capacities of *P. pullulans*, (but see the description of the fungus in the book by JÖRGENSEN, HANSEN & LUND, 1939, where weak fermentation is considered normal), these workers restricted fermenting strains from this species and erected a new species for them. Depending on their ability to ferment mannose, glucose, fructose, maltose, sucrose, raffinose, xylose, galactose and rhamnose, six varieties were established. The "fermentation" found by WYNNE & GOTT is of the aerobic type. The fact that acids were produced in the media in which these strains were grown indicates to this writer only that the strains involved are unable to completely metabolize the sugars under the conditions of the experiment.

Some external pigment was produced by several of the strains used by WYNNE & GOTT (1956). This was produced in the vicinity of the mycelium on nutrient agar or under anaerobic conditions. CIFERRI, et al. (1956), report that pigment production varies with the strain used and the oxygen tension. LUTERAAN et al. (1947) suggest that these melanin pigments may participate in the respiration process.

Few records are available concerning the temperature requirements of this fungus. WYNNE & GOTT in Texas found that most strains grow at 27 °C, while CIFERRI, et al., (1956) in Italy found that room temperature was optimum temperature for growth while maximum temperature lay between 35° and 40° C. RENNERFELT (1941) in Sweden, reported that the best temperature for the growth of his strains was 27° C.

CIFERRI, et al. (1956) report that no strains of *Aureobasidium pullulans* are known in which there is fermentation with gas on any sugar. Acid production occurred regularly when the fungus was grown on glucose and mannose. Occasionally, acid production can be obtained on levulose, maltose, rhamnose, raffinose, lactose, xylose. arabinose, dextrin and inulin. However, this capacity may become lost with age or may become erratic, depending on as yet undefined conditions of nutrients and environment.

That morphological appearance is a result of physiological activity was shown in part by NICKERSON et al. (1956) by growing *P. pullulans* on glucose-glycine-yeast extract (GGY) agar. During the first 48 hours a colorless budding yeast phase developed with occasional limited hyphal forms. After that time, at the periphery of the colony hyphae developed which became dark with abundant pigmentation. On these hyphae blastospores were produced singly or in clusters. When .001 M selenite was added, no pigmentation or mycelium was formed, but a heavy colorless yeast developed. After one week the colony became colored pink to light red. Selenite had been accumulated and was being reduced to selenium. It was thus shown that selenite promotes cell division.

CLARK & WALLACE (1958a, b) in Canada, approached the problem of the physiology of *Pullularia pullulans* from the point of view of cellular metabolism. Their work on carbohydrate metabolism was followed by a report on oxidation of compounds in the Krebs cycle. In all experiments they used the same strain of fungus — A.T.C.C. No. 11942. The authors' abstracts of these two contributions will be used to report their findings.

"*Pullularia pullulans*, A.T.C.C. No. 11942, has constitutive enzymes for the utilization of galactose, glucose, maltose, mannose, raffinose, sucrose and trehalose, but not for lactose. The organism oxidized mannose more rapidly, and maltose and raffinose more slowly, than any of the other sugars. Moreover, it utilized galactose and glucose at approximately the same rate, but more rapidly than trehalose; the oxidation of sucrose, however, was very slow at first and then very rapid. When resting cells were incubated with glucose-1-C^{14}, the carbon dioxide immediately produced was highly radioactive; when glucose-6-C^{14} was used, however, a considerable time elapsed before the production of noticeable amounts of radioactive carbon dioxide. The ratio of the activity of carbon dioxide produced from glucose-l-C^{14} to that produced from glucose-6-C^{14} was greater in young cells than in old. The results indicated that resting cells catabolize glucose in such a manner that most of the carbon dioxide comes from carbon-l of glucose. Extracts from cells were found to have an active phosphoglucomutase, glucose-6-phosphate dehydrogenase, 3-phosphoglyceraldehyde dehydrogenase, and fructose diphosphate aldolase. Both dehydrogenases were TPN linked. The extracts also oxidized ribose-5-phosphate but not ribose. The organism has enzymes, therefore, to catabolize glucose by some of the reactions of the Embden-Meyerhof system as well as by those of the pentose phosphate cycle."

"Studies with intact cells and cell free extracts of *P. pullulans* showed that the organism has the necessary enzymes for oxidizing most of the compounds of the Krebs cycle. The rates of oxidation of substrates by intact cells were studied in relation to the age of the cells, the concentration of substrate, the concentration of buffer, and the pH of the reaction medium. Cells grown for 36 hours oxidized a greater number of substrates than those grown in greater or less intervals. Cells grown for 6 and 18 hours had adaptive enzymes for the oxidation of acetate. The respiratory activity of the 36-hour old cells was generally greater in 0.0143 M concentration of substrate than in lower or higher concentrations; it was not affected by changes in concentration of buffer. With cells at pH 6.8, acetate and ethyl alcohol were oxidized at rates of more than one-half and one-third, respectively, that of glucose; alpha-ketoglutarate, fumarate, *cis*- aconitate, and succinate were slowly oxidized; and citrate, isocitrate, and malate were not oxidized. At pH 3.0, the cells oxidized most of the substrates more rapidly than they did at pH 6.8, but they did not oxidize citrate, *cis*-aconitate, nor isocitrate. The organism oxidized malonate, and its enzymatic activity apparently was not inhibited by that compound."

"Cell extracts oxidized alpha-ketoglutarate, citrate, fumarate, malate and succinate. TPN stimulated the oxidation of citrate and the activity of isocitrate dehydrogenase. DPN stimulated the oxidation of malate. Moreover, the activity of succinate dehydrogenase was inhibited by malonate. The results suggest that the metabolism of *P. pullulans* is by means of the reactions of the Krebs Cycle."

Among other fungi, BERNIER (1958) used *P. pullulans* in an investigation of polysaccharide production by litter and decay fungi growing on aqueous extracts of fresh leaves or litter, and on a glucose-salts-yeast extract solution. Sugar constituents of the polysaccharides of mycelial material of *P. pullulans* included glucose, mannose, traces of galactose and rhamnose, and glucuronic acid. *P. pullulans* produced polysaccharides on aqueous extracts of fresh leaves or litter of *Calluna*, beech, w. elm, ash and hazel. In many cases this was without addition of glucose to bring the glucose to a final concentration of 5 %.

RELATIONSHIPS WITH OTHER FUNGI

The polymorphic nature of *A. pullulans* has been indicated in the description of its morphology. That it can be and has been confused with other fungi is clearly indicated in the extensive synonymy. BERLÈSE (1895) said: "D'après les études faites, il me semble cependant qu'on puisse plus sûrement affirmer que *Dematium pullulans* est une forme collective, un état spécial d'un certain nombre de champignons non seulement sphériacés mais appartenant aussi à d'autres groups."

The picture is not as gloomy as BERLÈSE painted it. However, until, through cultural techniques, the fungus of which a *Pullularia*-like culture is a stage is determined, or during the time a known culture is being studied in the laboratory in its *Pullularia*-like stage, the critical student must be patient.

The fungus with which it is most commonly confused is *Cladosporium*. This is because the thick-walled brown cells, which occur in simple short chains or in long uninterrupted chains in the mycelium, may break up into arthrospore-like cells or into single chlamydospores. When such cells have a single septum they appear very much like spores of *Cladosporium*, which, by definition, has brown, thick-walled, two-celled spores, at least in some of its species. In primary isolation work, especially in areas where both *Cladosporium herbarum*-like species and *A. pullulans* are common members of a population, further confusion may occur when both fungi are isolated on the same plate and colonies appear so close together that mixed cultures of both species are obtained.

JANCZEWSKI (1892) considered *D. pullulans* as the first step in the development of *Cladosporium cladosporioides* in which *Hormodendrum cladosporioides* was an intermediate developmental stage. Later stages in the development of this series passed through *Phoma* to *Septoria* and *Leptosphaeria*. FERRARI (1932) reduced *Cryptococcus metaniger* to synonymy with *Cladosporium*. He amended *Cryptococcus* to exclude *Cladosporium* but to include *P. pullulans*. By 1956, CIFERRI had concluded that *C. metanigrum* was only a growth phase of *A. pullulans*. BRETT (1948) continued the discussion of the relationship between *Pullularia*, *Hormodendrum* and *Cladosporium*. While the latter two genera receive most space, it is indicated that there is a sequential relationship between these three genera.

SACCARDO'S (1878) *Tubercularia ampelophila* was associated with *Gloeosporium ampelophagum* fruiting on grapes. No reports in more recent literature repeat this observation. VON ARX (1957) lists the *Gloeosporium* with *Sphaceloma* and makes no mention of the *Tubercularia*. However, he recognizes three genera in which spores are produced as in *Aureobasidium*: The other two are *Kabatiella* and *Microstroma*.

In his definition of species in these genera, LINDAU (1905) indicated that confusion could arise, but that the two were quite distinct. As early as 1888, LAURENT indicated that the two were distinct. In his monographic treatment of *Cladosporium*, DE VRIES (1952) also indicated that the two genera could be easily confused. *Cladosporium werneckii* HORTA was based on a culture of a fungus obtained in Brazil from a case of Tinea nigra. In culture, this fungus has features which are similar to those of *Pullularia*, and DE VRIES placed it in that genus as a second species. While CIFERRI et al. (1956) considered this identical with *Aureobasidium pullulans*, CASTELLANI & DE SILVA (1957) have returned it to *Cladosporium* as a synonym of *C. mansoni*, a species first isolated from a case of Tinea nigra in Ceylon. The situation thus appears to be deadlocked, its clarification depending on an extensive cultural study of a large number of cultures from many areas and habitats.

HENRICI, in SKINNER, EMMONS & TSUCHIYA (1947), and discussions in the same volume by SKINNER, tend to consider the fungus, or forms similar to it, related to, if not identical with *Cladosporium.* It is shown that the fungus is initially yeast-like in culture, later developing a mycelium with budding conidia much like a *Candida,* and still later producing dry, aerial, conidia-bearing hyphae. In late stages, chains of two-celled, *Cladosporium*-like, "Spores" are produced. It is possible that here we have a pleomorphic or polymorphic fungus which in some strains, on some media, or under as yet undefined cultural conditions, is able to diverge in development simulating several genera of Fungi Imperfecti with none of which is it closely related.

SCHOSTAKOWITSCH (1895) has shown that the tips of the hyphal branches of the coremium-like perithecial initial of *Fumago* are conidiophores which, when separated from the parent hypha, can be mistaken for segments of *Dematium pullulans* sporulating hyphae. He indicated, however, that the relationship of *D. pullulans* is with *Hormodendrum* and *Cladosporium,* rather than with *Sphaerulina.*

BREFELD (1891) described and fully illustrated the fungus *Sphaerulina intermixta* showing that the spores of this species germinate as do the *Coniothecium-* like hyphal masses of *D. pullulans.* From this he reasoned that *D. pullulans* is the imperfect state of *Sphaerulina intermixta.* A number of workers, mostly within Germany, and as recently as NIETHAMMER (1937, 1947) have followed him in this interpretation. However, WEHMEYER (1957) has shown that *S. intermixta* is probably the same as *Saccothecium sepincola* (FR.) FR. and in her work with the anthracnose of roses JENKINS (1932) found that she could not demonstrate a connection between *Pringsheimia sepincola* (FR.) HÖHN and *D. pullulans.*

In 1929 MARTIN pointed out that in their developmental phases many genera of fungi parallel each other and thus may be confused one with the other. The phase in which MARTIN was especially interested was one which he called the "pseudosaccharomyces" or "pseudofumago" phase. It is characterized by the presence of indistinguishable budding or fumagoid cells in the mycelium. Representatives of the following genera which possessed such a phase were isolated from 50 host plants and were studied in pure culture: *Dematium pullulans* DE BARY, *Kabatiella* VASIL., *Polyspora* LAFFERTY, *Exobasidiopsis* KARAK., *Pachybasidiella* BUB. & SYD., *Aureobasidium* VIALA & BOYER, *Fumago vagans* PERS., *Protocoronospora* ATK. & EDG., *Microstroma* NIESSL and *Sphaceloma* DE BARY. It will be noted by reference to the synonymy of *A. pullulans* that most of these genera are considered synonyms of *Aureobasidium.* MARTIN considered these fungi as separate and distinct entities. He listed a number of genera of unrelated fungi as producing *Pseudosaccomyces* or *Pseudofumago* stages during culture on one or another type of medium in the laboratory. No doubt anyone who has worked with plant pathogenic fungi in pure culture can increase the list.

That several different types of fungi may have *Aureobasidium*-like stages in their developmental pattern is indicated by the fact that GROVES & LEACH (1949) reported that in three species of *Tympanus* studied in pure culture, the initial development phase resembled a culture of *Pullularia*. DELITSCH (1943) cited *Sphaerulina intermixta* and *Dothidea ribesia* as fungi with *Pullularia*-like imperfect states. PLANCHON (1900) also includes *Nectria cinnabarina, Rosellinia velutina* and *Bulgaria inquinans* in this list. KLEBAHN (1924) thought that the imperfect fungus arising from pure cultures of *Guignardia niesslii* KUNZE on *Iris* resembled *D. pullulans*, but that fungus was really a *Sporotrichum*. According to GÄUMANN (1952) the genera *Capnodium, Teichospora* and *Limacinia* have imperfect states which have been described as similar to cultures of this fungus.

The form of this fungus found by COSTANTIN & DUFOUR (1921) in the flowers of *Monotropa*, and which they called *Monotropomyces*, was suggested by them to be a stage of the *Phoma* or *Phyllophoma* thought to be the mycorrhizal symbiont of this and other ericaceous plants, although no phomaceous fungus resulted from cultures they prepared.

In his book on stem and leaf fungi, GROVES (1937) described *Polyspora* accurately, but associated it with the genus *Rhodesia*, both genera being considered melanconiaceous and not true Coelomycetes.

BENNETT (1928) thought that he had obtained the perfect state of *Pullularia pullulans* in his work with an *Anthostomella* which he called *A. pullulans*. VIENNOT-BOURGIN (1950) accepted BENNETT'S work, listing *A. pullulans* as the cause of grape leaf scald and giving genera such as *Dematium, Aureobasidium, Exobasidium* and *Pullularia* as synonyms. NANNFELDT (1934) questioned BENNETT'S conclusions since he had studied many strains of *Pullularia* from various stages in the process of paper making in Sweden, and had tried to obtain pairings between these strains and those of BENNETT without success. In NANNFELDT'S discussion of this problem, suggested techniques and related genera are indicated for the information of those who wish to try to obtain a perfect state.

That the fungus is highly polymorphic is shown by work in England by HOGGAN (1924) who suggested on the basis of his experiments that strains of fungi which had been referred to *Dematium pullulans* were of different unrelated fungi.

ROBAK (1952) noted that pycnidiospores of *Dothichiza pithyophila* (CHR.) PETR. (*Phomopsis pithyophila* CHR.) produced mycelium typical of *Pullularia*, but no mention was made of obtaining *Phomopsis* pycnidia on *Pullularia* mycelium.

At this point it may be mentioned that the only cytologic study of this fungus found was that of ROUKHELMAN (1937) in studies of *Torula schoenii*. It was found that the pigment was first formed in the vacuoles, then became deposited in the cell walls. Following the staining of the yeast-like conidia in iron hematoxylin, 2—3 nuclei per cell were noted, with some cells showing as many as 5 nuclei. This heterocaryotic condition may help to explain some of the extreme variability of this fungus. It is not improbable that the entity being discussed under the name of *Aureobasidium pullulans*, may in reality be the imperfect state of a number of different species of Ascomycetes.

Working with the related species *Kabatiella caulivora*, cause of an anthracnose disease of clover, COLE & COUCH (1959) found that the conidial cells contained 1—8 nuclei. In 100 spores there was a mean of 2.8 nuclei, 2 nuclei occurred in 36—38 %, and 8 in 1—2 % of the spores. The chromosome number was probably 4 although clumping resulting from fixing made counting difficult. These writers found no evidence for the dual phenomenon in this fungus. The mean nuclear number for cells of each growth type was the same. The mycelial or (M) type was stable, the conidial or (C) type of growth was unstable and the intermediate or (CM) growth type was absent. Starting with 500 individual conidia resulting from colonies isolated from a host plant, 2.4 % produced stable (M) colonies. Of the colonies developed from the remaining 488 conidia, 10 were chosen for further study. Five hundred conidia from each of these colonies were grown and 0.94 % produced (M) colonies the remainder being (C) colonies. In a second series grown from these (C) colonies 0.34 % were (M)-type colonies, the remainder (C)-type colonies.

EFFECT OF ANTIBIOTICS, CHEMOTHERAPY, AND FUMIGANTS

Aureobasidium pullulans, the black yeast, is important in medical agricultural, soils and industrial mycology, yet its activity in the presence of various chemotherapeutants, soil fumigants and antibiotics has been considered of interest partly because of its occurrence as a contaminant, partly because of its availability as a test organism.

In assaying the antifungal properties of culture solutions in which *Chaetomium globosum* had been grown, CUMMINGS (1954) found that while good growth (++++) was obtained in the control solution, *Pullularia* produced only fair (++) growth in media containing growth products of *Chaetomium*.

Ultra violet light can be used to control the growth of most organisms. HOLLAENDER (1942) reported that the greatest sensibility of "fungus" cells was approximately 2650 Angstrom units, close to that of the absorption of ultra violet light by nucleic acids. ENGLISH & GERHARDT (1946) had no success when they attempted to control the incidence of fungi which cause fruit rots in storage and transit. They used 30 Watt UV commercial germicidal lamps giving 2537 Angstrom units. In Petri dish exposures, 24.7 % of the spores tested were killed after 0.5 minutes, 28.7 % after 1.0

min., 58.3 % after 2.5 min., and 70.2 % after 5.0 min. After 45 minutes of irradiation with commercial equipment, the fungus was still isolated from sections of conveyor belts which carry fruit. The natural decay, caused by 5 fungal species including *Pullularia*, of sweet cherries was not affected by UV radiation even after exposure to doses up to 24 times as much as usually applied. Using daily exposures with low intensities of UV light, DILLON-WESTON & HOLMAN (1930) were only able to modify the rate of growth of the mycelium of this fungus. SCHNEGG, et al. (1936), have tried to control potentially harmful organisms in the brewery by the use of UV light. They used a Hanau-Uster lamp, added various concentrations of inoculum to water, irradiated the water and counted the number of survivors. Slime formers including *Dematium pullulans* were highly resistant in the UV treatment but these organisms were less dangerous than others in the beer. The irradiated water was useful in the brewery since no deleterious effects were noted on yeasts or on the fermentation process.

WIKÉN (1946) tested extracts from sporophores of 57 different mushroom-type fungi from Sweden for antibiotic activity against *P. pullulans*. *Boletus bovinus* and *Gomphidius glutinosus* produced the most powerful and persistent antagonism of this group of fungi. Twenty-one other species produced measurable inhibition zones which were only temporary. BRIAN & HENNING (1947) in England tested 115 fungi for antibacterial or antibiotic activity. Neither a strain called *Pullularia nigra* nor one labeled *P. pullulans* produced any such activity.

In a search for an antifungal antibiotic, *P. pullulans* was used among other fungi in testing the antifungal nature of candicidin (LECHAVALIER, et al., 1953). One to three μg/ml candicidin was needed for complete inhibition of this fungus.

In southern California it has been found that a large population of fungi has been built up in the soil of citrus orchards. Some of these fungi cause damage to the trees, and one way to prevent this is to sterilize or fumigate the soil. A population of soil microorganisms then may be developed which does not include the harmful species. One of the commoner fungi in such soils is *Pullularia pullulans* which has been shown (MARTIN, 1950) to cause little if any damage to orange seedlings and roots. This fungus is killed by the use of steam under pressure. However, if ethylene bromide, CS_2, or ethylene dichloride are used the fungus will recover after 2—8 months; and some recovery will be effected after 6 months when the soil is treated with dithane and with hexachlorocyclohexane and sour oranges are planted in it. Water and alcohol are the only leaching agents not affecting *Pullularia*. In 1957, MARTIN et al. reported that *Pullularia pullulans* returned to some soils as the dominant fungus after fumigation with D-D, CS_2, chloropicrin, vapam, and ethylene dichloride.

In the isolation of fungi from soil samples it has been found

(Martin, 1950c; Martin & Harding, 1951) that the use of rose bengal and streptomycin in primary isolation media reduces the numbers of bacteria and increases the numbers and kinds of fungi recoverable from soil samples. By reducing bacterial competition and bringing cultural conditions nearer to optimum conditions for more species of fungi, this medium has permitted the observation of many species of fungi not previously found or thought to be important members of a population. One of the fungi found by this technique to be a common member of the soil population is *Pullularia pullulans*. By using modifications of this medium, Cooke (1957, 1954b) was able to find this fungus as a common inhabitant of trickling filter slimes and a common member of the populations of various phases of sewage treatment. It was also found to be relatively common in a polluted stream.

That this fungus was able to produce its own antibiotic effects has been shown by Jeffreys et al. (1953) in England. They used three strains in streak culture tests and one in liquid culture. All streak cultures were shown to have antibiotic effects against the test organisms, and the filtrate of the liquid culture was shown to be inhibiting to *Bacillus subtilis* and slightly inhibiting to *Botrytis* sp.

In Finland a satisfactory method for using chloramine had been developed as a disinfectant for wood pulp, although in Sweden it gave conflicting results. Schmid (1935) found that a method patented in Norway was satisfactory. 90—95 % by weight of chlornaphthalene and 5—10 % mercury acetate were heated until most of the acetic acid had evaporated. The resultant transparent liquid was sprayed over the pulp or cellulose at a rate of 10—100 gm. per ton.

To prevent mold growth in wooden saranac container-type butter boxes, Riddett & Mill (1936) in New Zealand found that submersion of the wood in 0.1 % sodium salicylanilide (Shirlan W.S.) gave promising results. Sandborn (1937), in New York, suggested the chlorination of process water and the addition of copper sulphate at certain steps in the manufacture of paper would cut down the amount of slime growth which accompanies paper manufacture.

England (1939) in Sweden found that among the more efficient chemical sterilizers of wood pulp were the chlorphenols Dowicide and Santobrite. Sulphuric acid was also good. Rennerfelt (1941) reported that ethyl mercury chloride (pulposan) was a good antiseptic, and that Santobrite, Dowicide, and Tebecit (sodium orthophenyl-phenolate) used at the rate of 10, 40 and 100 times, respectively, as much as the appropriate dosage of pulposan (200 g/Ton) were good controls for *P. pullulans* in the paper mill.

Van Luyk (1938) in Holland, found that a strain of *P. pullulans* produced a substance antagonistic to five species of *Phythium* tested alone and in mixed culture. This substance was stable at 100° C.

In work with pears, ENGLISH (1940) found that high temperatures 32—35° C, rather than common storage, room, and cold temperatures, tended to inhibit growth of most of the fungi associated with decay during storage and transit. In work with sweet cherries, ENGLISH & GERHARDT (1943) noted that decay fungi can be controlled with concentrations of CO_2 in the range used in commercial shipment practices.

In New Zealand (Anon., 1940) it has been found that oil flax seed, of the variety Rio, could be treated satisfactorily for the control of *Pullularia* by immersion for 10 min., in water heated to 126° F.

MUSKETT & COLHOUN (1941) in Northern Ireland recommended the use of RD 7846 — methylthiuram disulphide (Nomersan) for seed treatment of flax. Two thousand tons of seed were so treated in 1941 with only occasional skin irritations by contact with the fungicide reported. Ceresan U-564 also gave good results using the short liquid method and the Koutrainix continuous seed treating apparatus. Either treatment resulted in good crops with no phytocidal effect.

Painted filter paper discs have been used (VICKLUND & MANOWITZ, 1951) as a means of testing efficacy of fungicides in paints against this fungus. On the basis of 7—14 days growth it was found that 0.5 % copper-8-quinolineolate had outstanding fungicidal activity. Since mildew resistance is a function of the surface concentration of fungicides, the use of fungicides is necessary even when zinc-oxide is present.

In Sweden, PEHRSON (1947) found a bacterium in mechanical pulp and white water of which 5 of 7 strains produced an antibiotic effective against *P. pullulans*. A 1 ml. suspension of fresh mechanical pulp and white water was placed in a Petri dish with 1 ml spore suspension, wort agar was poured into the dish, and the whole incubated at 35° C for 24 hours, then after inverting, incubating at 23° C for 3 days. Clear zones developed around dark fungal colonies, the organism was reisolated and retested. The inhibition zone around the fungus measured 19—21 mm. by the Oxford test, or 20—25 mm. by the streak test. The substance was thermostable at 121° C, some loss of activity was shown at 4° C, and at a pH of 11 a noticeable decline was shown.

RENNERFELT (1949) has found that organic mercury compounds and certain chlorophenols are the most effective antiseptics in treating against bluing and decay in wet mechanical pulp in Sweden.

While fire resistance-treated cotton duck resisted fungal attack (MELORO, 1950) when *P. pullulans* was used as a test organism, certain copper compounds had varying degrees of inhibitive action. One-half per cent copper naphthenate or hydroxynaphthenate did not inhibit growth alone. However, when 0.2 % copper containing copper-8-quinolinolate (Milmer I) was used alone or in a concentration of 0.05 % copper with copper hydroxynaphthenate (0.385 % copper) fungal attack was successfully halted.

In England, BAILLIE et al. (1950) reported that Benztropone and α-thuyaplicin at 50 mg/l were fungicidal to *P. pullulans* and that no spores survived after 53 hours after exposure.

A test in Germany (SCHULZ, 1951) has shown that for inhibition of *Pullularia pullulans* as a cause of blue stain of pine wood, 1 % sodium pentachlorophenolate and 1.5 % of "protectant X" were required. The experiment was carried out in pure culture where blocks of wood 1.5 × 2.5 × 5.0 cm, soaked in 1 % malt extract and varying percentages of the protectants were incubated for 4 weeks in cotton stoppered flasks.

Experiments in Finland (GADD 1951) have shown that bluing of paper pulp occurs even though pulp is kept at below 25 % moisture and in cultures fungal growth is inhibited below 30 % moisture. Optimum temperature for growth of bluing fungi is between 45—60° F, depending upon species and conditions involved. It was found that 20—25 g/wet ton of phenylmercuric acetate is effective in arresting fungal deterioration.

To protect paper used in archives for permanent records, KOWALIK (1952) found that the best protection against *Pullularia pullulans* was paper made from 95 % alpha-cellulose with 1 % copper compound and pH of 5.5—6.0. Zinc-dimethyl-dithiocarbamate added during production was also suggested.

GARINO-CANINA (1929) noted that no growth of this fungus occurred in a grape must medium to which had been added 0.179 g of SO_2/l.

Sorbic acid is becoming a popular fungistatic agent for use in various types of foods (MELNICK et al., 1956). BENEKE & FABIAN (1955) used it in a puree of thawed frozen strawberries. While *P. pullulans* grew well on control puree to which nothing had been added at pH levels of 3.3 and 4.0, when sorbic acid was added to either pH level at concentrations of as low as 0.025 %, no growth occurred in inoculated plates.

When this fungus appears as the causal agent of Tinea nigra, several types of remedies can be used in stopping the spread of the disease, according to CASTELLANI & DE SILVA (1957). In early stages several successive daily applications of formalin can be used. In older cases a salycilic alcohol lotion followed by resorcin ointment is effective. Among other antimycotic salves, a salycilic sulphur ointment may be prescribed. Tincture of iodine and other iodine-containing liquid preparations have been used. CASTELLANI'S fuchsin paint is recommended in Ceylon and elsewhere.

SURVIVAL AND LONGEVITY

No tests have been designed to determine the length of time during which *Aureobasidium pullulans* can survive preservation in the dry condition. However, in a test of survival of cultures of fungi kept under mineral oil, it has been found (STEBBINS & ROBBINS, 1949) that *P. pullulans* has survived for one and two year periods. Reports of survival after lyophilization, freezing and soil burial have not included this fungus in the lists of species studied. No information is available concerning length of survival of the fungus after prolonged periods of desiccation in nature or under conditions of storage of herbarium or culture collections.

SUMMARY AND CONCLUSIONS

Aureobasidium pullulans (DE BARY) ARNAUD, as understood in this report, is a common, ubiquitous, omnivorous, and highly polymorphic species of fungus. Since the cells of at least one, and possibly most, strains contain two to five nuclei it is presumed to be heterocaryotic. Since its spores are wind-borne, mycelia developing in one area may have germinated from spores originating in many areas. Processes of mitotic gene exchange known to occur in heterocaryotic fungi can thus contribute to the morphological and physiological variation found in this fungus. Surveys of culture conditions, nutrient requirements and morphological variation, could well be developed using stock culture strains as well as strains isolated from as many natural situations as practical to determine the extent of variation possible in this fungus and the cultural properties of strains known to be uninucleate.

A further consideration has come to light with the publication of a revision of the generic affiliations of the species of a group of plant pathogens. Questions concerning relationships between weak plant pathogens of greater or lesser importance and saprobes of extensive distribution need clarification.

RÉSUMÉ ET CONCLUSIONS

Tel qu'il apparaît dans le présent compte-rendu de recherches, l'*Aureobasidium pullulans* (DE BARY) ARNAUD est une espèce de champignon commun, omnipresent, omnivore, et à un haut degré polymorphique. Les cellules d'une lignée — et peut-être de la plupart des lignées — contenant de deux à cinq noyaux, cette espèce doit être présumée hétérocaryotique. Ses spores étant portées par les vents, les myceliums qu'elles développent dans une région donnée peuvent avoir pullulé à partir de spores produites dans un grand nombre de régions. Les procédés d'échange mitotique des gènes qu'on peut constater chez les champignons hétérocaryotiques peuvent ainsi contribuer aux variations morphologiques et physiologiques qui se rencontrent dans ce champignon. Une étude des conditions de culture, des nécessités d'ordre nutritif, et des variations morphologiques pourrait être poursuivie avec avantage, en utilisant des lignées de culture souches, aussi bien que des lignées

qu'on aurait développées à l'écart d'autant de diverses conditions naturelles qu'il serait pratique de le faire; on pourrait ainsi déterminer l'importance des variations possibles dans ce champignon, ainsi que les propriétés culturelles des lignées reconnues comme étant uninucléées.

Une considération supplémentaire a été mise en lumière par la publication d'un recensement des affiliations génériques des différentes espèces, à l'intérieur d'un groupe de champignons pathogènes. Il y aurait lieu de clarifier les questions de relations entre champignons, agents pathogènes faibles, et champignons saprobiques de large répartition géographique.

BIBLIOGRAPHY

Adam, D. B. (1923) J. Dept. Agr., Victoria **21**, 307—317.

Airola, E. V. (1941) Papp. Tavaratidskr. Finl. **23**, 112—114, 116—118, 120—122, 124, 126, 128—129.

Alwood, W. A. (1908) Bur. Chem. U.S.D.A. Bull. **111**, 1—28.

Anon. (1925) Nature **116**, 187.

Anon. (1930) Rept. Calif. Agr. Exp. Sta. July 1, 1928 — June 30, 1929, 123 pp.

Anon. (1940) Ann. Rep. Dept. Sci. Ind. Res., N. Zealand, **14**, 1—100.

Anon. (1945) Trop. Det. Res. Lab. Phila., Oct. 1945.

Anon. (1945) Trop. Det. Res. Lab. Phila. Week Ending 6 Oct. 1945.

Arnaud, G. (1910) Ann. Mycol. **8**, 470—476.

Arnaud, G. (1931) Traité de Pathologie Végétal, Paris, Vol. 1, 2.

Arx, J. A. von (1957) Verh. K. Nederl. Akad. v. Wetensch., Afd. Natuurk. Tweede Reeks, **51(3)**, 1—153.

Bade, O. & P. Klem (1935) Papirjournalen **23**, 39—44, 47—49.

Baillie, A. J., G. C. Freeman, J. W. Cook & A. R. Somerville (1950) Nature **166**, 65.

Barghoorn, E. S. (1945) Interim Report, 30 June.

Batko, S., J. S. Murray & T. R. Peace (1958) Trans. Brit. mycol. Soc. **41**, 126—128.

Bauer, R. (1938) Zbl. Bakt. **II. 98**, 133—167.

Baylis, G. T. S. (1941) N. Zeal. J. Sci. Tech. **A. 23**, 1—8.

Beauverie, J. (1910) Ann. Sci. Nat., Bot. **IX**, 11, 31—73.

Becker, J. G. & C. G. Shaw (1955) Appl. Microbiol. **3**, 173—180.

Benedek, T. & G. Specht (1933) Zbl. Bakt. P. **I. 130**, 74—90.

Beneke, E. S. & F. W. Fabian (1955) Food Technol. **9**, 486—488.

Beneke, E. S., L. S. White & F. W. Fabian (1954) Appl. Microbiol. **2**, 253—258.

Bennet, F. T. (1928) Ann. appl. Biol. **15**, 371—391.

Berkhout, M. (1923) De Schimmelgeschlachten Monilia, Oidium, Oospora en Torula, Scheveningen, Holland.

Berlèse, A. (1906) Redia **3**, 8—15.

Bernier, B. (1958) Canad. J. Microbiol. **4**, 195—204.

Birkinshaw, J. H. (1937) Biol. Rev. **12**, 357—392.

Björkman, E. (1946) Statens Skogsförs. Medd., Sweden **35**, 5, 1—7.

Björkman, E. (1946) Statens Skogsförs. Medd., Sweden **35**, 7, 1—46.

Björkman, E. (1947) Svensk Papp. Tidn. **1** (11) B, 49—52.

Björling, K. (1937) Svenska Skogsy. Fören. Tidskr. **25**, 250—258.

Bolaunova, O. (1935) Pl. Prot. Leningrad 1935, **6**, 107—108.

Bouthilet, R. J. (1951) Mycopathol. Mycol. Appl. **6**, 79—85.

Brain C. K. (1923) Ann. Univ. Stellenbosch **1**, A, 11, 1—48.

Breest, F. (1914) Arch. Protistenk. **34**, 263—276.

Brefeld, O. (1891) Untersuchungen **10**, 266—267.

Brett, M. A. (1948) Trans. Brit. mycol. Soc. **30**, 141—151.

Brian, P. W. & H. G. Henning (1947) J. gen. Microbiol. **1**, 153—167.

Brooks, F. T. & C. G. Hansford (1923) Trans. Brit. mycol. Soc. **8**, 113—142.

Brues, C. T. & R. W. Glaser (1921) Biol. Bull. **40,** 299—324.

Buchner, P. (1911) S. B. Ges. Morph. Physiol. (München) **27,** 89—96.

Buchner, P. (1912) Arch. Protistenk. **26,** 1—116.

Buchner, P. (1912) Tier und Pflanze in intracellularer Symbiose, Berlin.

Buchner, P. (1953) Endosymbiose der Tiere mit Pflanzlichen Mikroorganismen. Stuttgart.

Burgess, R. (1924) J. Inst. Text. **15,** T573—T583.

Capriotti, A. (1956) Atti Acad. It. Vite e Vino, 8.

Carrion, A. L. (1950) Arch. Dermat. Syph. **61,** 996—1009.

Castellani, A. & A. J. Chalmers (1913) Manual of Tropical Medicine, London.

Castellani, A. & R. de Silva (1957) Mykosen **1,** 41—49.

Chesters, C. G. C. (1948) Trans. Brit. mycol. Soc. **30,** 100—117.

Christensen, C. M. & M. A. Swaebly (1951) Mimeo Contrib. Sect. Pl. Path. U. Minn.

Chrzaszcz, T. & R. Shillak (1936) Biochem. Z. **288,** 359—368.

Ciferri, R. (1939) Agricoltura Colon. **33,** 571—2.

Ciferri, R. & B. K. Ashford (1929) Puerto Rico J. Bull. Health and Trop. Med. **5,** 188—195.

Ciferri, R. & P. Redaelli (1957) Mycopathol. Mycol. Appl. **8,** 183—188.

Ciferri, R., M. Ribaldi & A. Corte (1956) Atti Ist. Bot. & Lab. Critt. Univ. Pavia **14,** 3—15.

Clark, D. S. & R. H. Wallace (1958a) Canad. J. Microbiol. **4,** 43—54.

Clark, D. S. & R. H. Wallace (1958b) Canad. J. Microbiol. **4,** 125—139.

Clements, F. E. & C. L. Shear (1931) The Genera of Fungi, New York.

Colhoun, J. & A. E. Muskett (1948) Ann. appl. Biol. **35,** 429—434.

Conant, N. F., et al. (1954) Manual of Clinical Mycology, 2nd ed., Philadelphia.

Cooke, W. B. (1951) Ecology **32,** 736—748.

Cooke, W. B. (1954) Sewage and Industrial Wastes **26,** 661—674.

Cooke, W. B. (1957) Sydowia, Ann. Mycol. Ser. II, Beiheft 1, 146—175.

Costantin, J. & L. Dufour (1921) C. R. Acad. Sci. Paris **173,** 957—959.

Coudert, J. (1955) Guide Pratique de Mycologie Medicale, Paris.

Cummins, J. (1954) Mycologia **46,** 289—292.

Cunningham, J. H. (1940) Pl. Dis. Div. Bull., N. Zealand **33,** 1—56.

Dale, G. (1914) Ann. Mycol. **12,** 33—62.

Davidson, R. W. (1935) J. agric. Res. **50,** 789—807.

Delitsch, H. (1943) Systematik der Schimmelpilze, Neudamm, Germany.

de Vries, G. A. (1952) Contribution to the Knowledge of the Genus Cladosporium. Link ex Fr., Baarn, Holland.

Diddens, H. A. & J. Lodder (1942) Die Anakosporogenen Hefen — Zweite Hälfte. Amsterdam.

Dillon-Weston, W. A. R. & E. T. Holman (1930) Phytopathology **20,** 959—965.

Dodge, C. W. (1934) Medical Mycology, St. Louis.

Doolittle, W. T. (1948) J. For. **46,** 928—929.

Dye, M. H. & T. R. Vernon (1952) N. Zeal. J. Sci. Tech. B. **34,** 118—127.

English, W. H. (1940) Taxonomic and pathogenicity studies on the fungi which cause decay of pears in Washington. Thesis. The State College of Washington, Pullman.

English, W. H. (1940) Res. Stud. St. Coll. Wash. **8,** 127—128.

English, W. H. (1945) Pl. Dis. Reptr. **29,** 559—566.

English, W. H. & F. Gerhardt (1943) Proc. Amer. Soc. Hort. Sci. **40,** 172—176.

English, W. H. & F. Gerhardt (1946) Phytopathology **36,** 100—111.

Englund, B. (1939) Zellstoff u. Papier **19,** 336, 338, 340, 342, 346, 348. Experiment Station Record (1894) **6,** 230—231.

Farrar, K. T. H. (1949) Aust. J. Sci. **12,** 31—32.

Ferrari, A. (1932) Atti Ist. Bot. R. Univ. Pavia **IV, 3,** 175—184.

Fisher, F. (1948) Fibers, Fabrics and Cordages **15,** 331—334.

Flor H. H. (1936) Phytopathology **26,** 93—94.

Flor, H. H. (1936) Phytopathology **26,** 429—438.

Foex, G. & P. Viala (1896) Rev. Vitic. **6,** 501—502.

Fraser, L. (1933) Proc. Linn. Soc. N. S. Wales **58,** 375—395.

Gabotto, L. (1907) Atti Congr. Nat. Ital., Milano, 514—521.

Gadd, G. O. (1950) Medd. Industrins Centrallab. Teollisunden Keskuslab. Tiedonantoja **102,** 49—52.

Galloway, M. A. (1930) Shirley Inst. Mem. **9,** 27—36.

Garino-Canina, E. (1929) Riv. Ampelograf. **1,** 2—6.

Gäumann, E. A. (1952) The Fungi, Translated by F. L. Wynd, New York.

Gilman, J. C. (1957) A Manual of Soil Fungi, 2nd. ed., Ames, Iowa.

Goll, M. & G. Coffey (1948) Paint, Oil Chem. Rev. **111** (16), 14, 16—17, 30.

Gray, W. D. (1945) Rept. 3, Eng. Div., Jeffersonville Q.M. Dept. Dec.

Groves, J. W. & A. M. Leach (1949) Mycologia **41,** 59—76.

Groves, W. B. (1937) British Stem and Leaf Fungi, Cambridge, Vol. II.

Gruenhagan, R. H., A. J. Riker & C. A. Richards (1947) Phytopathology **37,** 757—772.

Guilliermond, A. & F. W. Tanner (1920) The Yeasts. New York.

Haddow, W. R. (1941) Trans. Roy. Canad. Inst. **23, 161**—**189.**

Haensler, C. M. (1921) Ann. Rept. N. J. Agr. Exp. Sta. **41,** 605—607.

Haensler, C. M. (1921) N. J. Agr. **3,** 6, 4.

Hansam, W. (1936) Collegium **1936,** 561—566.

Hansam, W. & E. Liebscher (1940) Collegium **1940,** 506—508.

Henneberg, W. (1926) Handbuch der Gärungsbakteriologie, 2nd. ed. **2,** 308—309.

Herr, L. (1957) Ohio J. Sci. **57,** 203—211.

Hedgcock, G. G. (1906) Ann. Rept. Missouri Bot. Gard. **17,** 56—114.

Henderson, M. E. K. & V. E. Farmer (1955) J. gen. Microbiol. **12:** 37—46.

Hoggan, I. A. (1924) Trans. Brit. mycol. Soc. **9,** 100—107.

Hollaender, A. (1942) Aerobiology. Washington.

Hubert, E. E. (1929) Wood Utilization, U. S. Dept. Commerce, Washington.

Hubert E. E. (1938) Ind. & Eng. Chem. **30,** 1241—1250.

Hubert, E. E. (1940) Ind. & Eng. Chem., Analyst Ed., **42,** (139—141).

Hyde, E. O. C. (1938) N. Zeal. J. Agr. **57,** 301—302.

Janczewski (1892) Acad. Sci. Gracovie Bul. **10,** 417—422.

Jeffreys, E. G., et al. (1953) J. gen. Microbiol. **9,** 314—341.

Jenkins, A. E. (1932) J. agric. Res. **45,** 321—337.

Jensen, H. L. (1941) Aust. J. Sci. **4,** 59.

Jensen, H. L. (1941) Proc. Linn. Soc. N. S. Wales **66,** 276—286.

Jörgensen, A., E. C. Hansen & E. C. Lund (1939) Microorganism and Fermentation, London.

Jump, J. A. (1938) Phytopathology **27,** 798—811.

Kazina, O. (1935) Pl. Prot. Leningr. **1935,** 100—115.

Keener, P. D. (1950) Amer. J. Bot. **37,** 520—527.

Klebahn, H. (1924) Ber. dtsch. bot. Ges. **42,** 60—71.

Klocker, A. & H. Schöinning (1900) C. R. Lab. Carlsberg **5,** 47—57.

Konopacka, W. (1931) Choroby Roslin (Plant Diseases) Warsaw **1,** 2, —14.

Kowalik, R. (1952) Med. Dosiwiadczalna i Mikrobiol. **4,** 359—360.

Kozucharov, K. (1937) Sbornik Ceskoslov. Akad. Zemedelske **12,** 14—22.

Krause, R. L. (1948) For. Prod. Res. Soc. Preprint **29,** 1—8.

Lagerberg, T., G. Lundberg & E. Melin (1927) Skogsf. Tidskr., Stockholm **25,** 145—272, 561—691.

Langeron, M. & P. Guerra (1938) Ann. Parasit. hum. comp. **16,** 436.

Langeron, M. & R. V. Talice (1932) Ann. Parasit. hum. comp. **10,** 1—80.

Laurent, E. (1888) Ann. Inst. Pasteur **2,** 581.

Lechavalier, H., et al. (1953) Mycologia **45,** 166—171.

Lewis, C. E. (1912) Phytopathology **2,** 49—62.

Lindau, G. (1905—1907) Handbuch der Technischen Mykologie. Jena.

Lindau, G. (1907) Rabenhorst's Kryptogamenflora **1,** VIII. Leipzig.

Lindner, P. (1888) Wschr. Brauerei **5,** 290—291.

Lindner, P. (1905) Mikroscopische Betriebskontrolle in den Gärungsgewerben, Berlin.

Lindner, P. (1895) Zbl. Bakt. P. II, **1,** 782—787.

Lipman, C. B. (1911—1912) J. biol. Chem. **10,** 169—182.

Löhnis, F. & N. K. Pillai (1896) Zbl. Bakt. P. II, **20,** 781—799.

Loew, E. (1868) J. wiss. Bot. **6**, 467—477.

Luteraan P. J. (1954a) C. R. Acad. Sci. Paris **239**, 595—597.

Luteraan, P. J. (1954b) C. R. Acad. Sci. Paris **239**, 1406—1408.

Luteraan, P. J., M. F. Champeau & J. Choay (1947) C. R. Soc. Biol. **141**, 616—617.

Madihassan, S. (1928) Arch. Protistenk. **63**, 18—22.

Madihassan, S. (1929) Arch. Protistenk. **63**, 613—624.

Malm, M. (1941) Svensk. Papp. Tidsk. 44. 23, 520—529; 24, 554—557.

March, P. B. & K. Bollenbacker (1949) Textile Res. J. 19, 313—324.

March, P. B., K. Bollenbacker, M. L. Butler & K. B. Raper (1949) Textile Res. J. 19, 462—484.

Martin, G. H. (1929) Phytopathology **19**, 1117—1123.

Martin, J. P. (1950) Soil Sci. **69**, 107—122.

Martin, J. P. (1950b) Soil Sci. **69**, 215—232.

Martin, J. P. (1950c) Soil Sci. Soc. Amer. Proc. **14**, 184—188.

Martin, J. P., R. C. Baines & J. O. Ervin (1957) Soil Sci. Soc. Amer. Proc. **21**, 163—166.

Martin, J. P. & R. B. Harding (1951) Soil Sci. Soc. Amer. Proc. **15**, 159—162.

Martin, J. P., et al. (1956) Soil Sci. **81**, 259—267.

Martin, J. T., R. F. Batt & R. T. Burchill (1957) Nature **180**, 796—797.

Mason, E. W. (1937) I.M.I. Papers, Fasc. **3**, General Part, 90.

Massee, G. (1898) Kew Bull. 1898, 321.

Mathieu, L. (1929) Rev. Vitic. **70**, 345—349.

Maunsell, K. (1954) J. Larynology & Otology **68**, 765—775.

Maurizio, A. & W. Staub (1928) Zbl. Bakt. P. II, **75**, 375.

McAlpine, D. (1898) Additions to Fungi on the Vine, Agric. Dept. Victoria. **1**—54.

McKay, R. & J. B. Loughnane (1947) J. Dept. Agric. Eire **44**, 37—40.

Meier, K. & H. Schmidt (1952) Deut. Farben. Z. **6**, 87—92.

Melin, E. & J. A. Nannfeldt (1935) Svensk. Skogsf. Tidskr. **32**, 3—4, 397—616.

Mellicker, J. B. & M. W. Baker (1942) Ind. Eng. Chem., Anal. Ed. **14**, 305—308.

Melnick, D., H. W. Vahlteich & A. Hackett (1956) Food Res. **21**, 133—146.

Meloro, F. A. (1950) Res. Serv. Text. Rept. C. & P.-79-F. U.S.Q.M. Depot, Phila.

Metchnikoff (1884) Arch. path. Anat. Physiol. klin. Med. **96**, 177.

Miller, J. H., J. E. Giddens & A. A. Foster (1957) Mycologia **49**, 779—808.

Moll, F. (1930) Mitt. Ges. Vorratsschutz **6**, 15.

Molnar, A. C. (1958) Ann. Rept. For. Insect & Dis. Surv., Canada Dept. Agr. 1958, 84.

Molnar, A. C. (1959) Canad. J. Bot. (in press).

Moniez, R. (1887) Soc. zool. France Bul. **12**, 150—152.

Montemartini (1897) Atti Ist. Bot. R. Univ. Pavia **II, 5**, 70—77.

Montoya, A. & J. R. Florey (1898) Thèse Fac. Med. Paris **21**. 48.

Morrow, M. B. (1947) Ann. Allergy **5**, 442—454.

Morrow, M. B. & E. P. Lowe (1943) Mycologia **35**, 638—653.

Morrow, M. B., E. P. Lowe & H. E. Prince (1942) J. Allergy **13**, 215—226.

Mrak, E. & H. J. Phaff (1948) Ann. Rev. Microbiol. **2**, 1—46.

Muskett, A. E. (1948) Trans. Brit. mycol. Soc. **30**, 74—83.

Muskett, A. E. & E. L. Calvert (1940) Nature, London. **146**, 200—201.

Muskett, A. E. & J. Colhoun (1941) Nature, London. **148**, 198—199.

Muskett, A. E. & J. P. Malone (1941) Ann. appl. Biol. **28**, 8—13.

Neger, F. W. (1918) Flora **110**, 67—139.

Negroni, P. & I. Fischer (1942) Rev. Inst. Pact. "Dr. Carlos G. Malbran" **11**, 99—108.

Newhook, F. J. (1947) N. Zealand J. Sci. Tech. A, **29**, 44—48.

Nickerson, W. J., W. A. Taber & G. Falcone (1956) Canad. J. Microbiol. **2**: 575—584.

Niethammer, A. (1937) Die Mikroscopischen Boden-Pilze. The Hague.

Niethammer, A. (1941) Arch. Mikrobiol. **12**, 312—328.

Niethammer, A. (1942) Holz Roh-u.-Werkstoff **8**, 269—273.

Niethammer, A. (1947) Technische Mykologie, Stuttgart.

Njoku-Obi, A. N., J. V. Spencer, E. A. Sauter & M. W. Eklund (1957) Appl. Microbiol. **5**, 319—321.

NOBLE, M. (1939) Ann. appl. Biol. **26,** 630—633.
Nuodex Products Co. (1955) Nuodex Microbiol. Test Methods, Elizabeth, N. J.
OGILVIE, L. (1924) Trans. Brit. mycol. Soc. **9,** 167—182.
PADY, S. M. (1951) Canad. J. Bot. **29,** 46—56.
PADY, S. M. & L. KAPICA (1953) Canad. J. Bot. **31,** 309—323.
PADY, S. M. & L. KAPICA (1955) Mycologia **47,** 34—50.
PADY, S. M. & L. KAPICA (1956) Canad. J. Bot. **34,** 1—15.
PADY, S. M. & C. D. KELLY (1953) Canad. J. Bot. **31,** 107—122.
PAVLONSHIN, P. J. (1937) Pl. Prot., Leningr. **1937,** 34—43.
PEHRSON, S. (1947) Svensk. Bot. Tidskr. **42,** 354—364.
PHAFF, H. J., E. M. MRAK & O. B. WILLIAMS (1952) Mycologia **44,** 431—451.
PIERANTONI, U. (1925) Scientia, Milano. **37,** 85—92.
PLANCHON, L. (1900) Ann. Sci. Nat. **11,** 167—226.
POTEBNIA, G. (1897) Trav. Soc. Nat. Univ. Krakow **31,** 28.
PRIBRAM, E. (1925) Ergeb. Physiol. **24,** 95—106.
PRILLIEUX, E. (1895) Maladies de Plantes Agricoles, Vol. I. Paris.
PRILLIEUX, E. & G. DELACROIX (1894) C. R. Acad. Sci. Paris **119,** 106.
PRINDLE, P. (1935) Textile Res. **5,** 542—568; **6,** 23—43.
RAYBAUD, L. (1923) Congres Path. veg. Strasbourg 48—49.
REDAELLI, P. & R. CIFERRI (1947) Mycopathol. **4,** 25—47.
REESE, E. T., H. CRAVETZ & G. R. MANDELS (1955) Farlowia **4,** 409—421.
REESE, E. T. et al. (1950) Farlowia **4,** 45—86.
RENNERFELT, E. (1937) Svensk. Skogsv. Fören. Tidskr. **35,** 43—159.
RENNERFELT, E. (1939a) Svensk. Papp. Tidn. **42,** 2—5.
RENNERFELT, E. (1939b) World's Pap. Tra. Rev., Tech. Suppl. **112,** 169—175; **113,** 1—
RENNERFELT, E. (1939—1940) Medd. Göteborgs Botan. Tradgård. **13,** 1—10.
RENNERFELT, E. (1941a) Svensk. Skögsf. Tidskr. **39,** 19—94.
RENNERFELT, E. (1941b) Arch. Mikrobiol. **12,** 19—40.
RENNERFELT, E. (1946) Medd. Skögsf., Stockholm **34,** 391—416.
RENNERFELT, E. (1947) Svensk. Bot. Tidskr. **41,** 283—294.
RENNERFELT, E. (1949) Rep. Proc. 4th. Int. Congr. Microbiol. **1947,** 403—404.
REYNOLDS, E. S. (1950) Mycologia **42,** 432—448.
RICHARDS, C. W. (1949) J. Bact. **58,** 453—455.
RICHTER, H. (1941) Mitt. Ges. Vorratsschutz **7,** 2—5, 22—23.
RIDDETT, W. & J. C. MILL (1936) N. Zeal. J. Agr. **53,** 129—139.
ROBAK, H. (1932) Nyt. Mag. f. Natvsk. **71,** 185—330.
ROGERS, D. P. (1951) Letter to W. L. WHITE dated 5-25-51.
ROSA, D. G., E. B. FRED & W. H. PETERSON (1929) Zbl. Bakt. II, **79,** 86—92.
ROUKHELMAN, N. (1937) C. R. Soc. Biol. Paris **124,** 545—547.
SABET, Y. S. (1935) Egyptian Univ. Bull. Fac. Sci. **5,** 1—29.
SANDBORN, J. R. (1937) Ind. Eng. Chem. **29,** 949—951.
SCHAEFER, T. G. & H. S. CHICHESTER (1948) Stk. Lamherm. 15 Dec. 3 pp.
SCHANDERL, H. (1942) Wschr. Brau. **59,** 59—61.
SCHEFER, T. C. (1946) Spec. Release, U.S.D.A. Div. For. Path. **28,** 1—6.
SCHMID, W. (1945) Papierfabrikant **33,** 380—382, 387—389.
SCHNEGG, H., H. KIPPHAN & K. GRUNERT (1936) Z. Ger. Brauw. **59,** 21—31.
SCHÖNNING, H. & A. KLÖCKER (1900) C. R. Trav. Lab. Carlsberg. 5.
SCHOPFER, W. H. (1938) Ann. Inst. Pasteur **61,** 781—782.
SCHOSTAKOWITSCH, W. (1895) Flora **81,** 362—393.
SCHULZ, G. (1951) Angew. Bot. **26,** 42—54.
SÉE, P. (1917) C. R. Acad. Sci. Paris **164,** 230.
SKINNER, C. E., C. W. EMMONS & H. N. TSUCHIYA (1947) Henrici's Molds, Yeasts and Actinomycetes, 2nd. ed., New York.
SMIT, J. & K. T. WIERINGA (1953) Nature, London. **171,** 794.
SMITH, R. E. (1929) Phytopathology **19,** 1139.
STEBBINS, M. E. & W. J. ROBBINS (1949) Mycologia **41,** 632—636.
STEINHAUS, E. A. (1947) Insect Microbiology, Ithaca.
STEINHAUS, E. A. (1951) Hilgardia **20,** 629—678.
STEINHAUS, E. A. (1955) Hilgardia **24,** 185—206.

STURGIS, W. C. (1901) Ann. Rep. Conn. Agr. Exp. Sta., New Haven 1900. **24,** 255—297.
ŠULC, K. (1906) S. B. Kgl. Böhm. Ges. Wiss. Art. **19,** 6.
ŠULC, K. (1910) S. B. Kgl. Böhm. Ges. Wiss. Art. **3,** 1—39.
ŠULC, K. (1910) S. B. Kgl. Böhm. Ges. Wiss. Art. **14,** 1—6.
SUMIKI, Y. (1929) J. agric. Chem. Soc. Japan **5,** 576—580.
SVOLBA, F. (1932) Gartenbau Wss. **7,** 282—292.
TAYLOR, C. F. & L. SHANOR (1945) Phytopathology **35,** 210—212.
TENGWALL, T. A. (1924) Meded. Phytopath. Lab. Baarn. **6,** 34—35.
TEODORO, G. (1918) Redia **13,** 1—5.
THAYSEN, A. C. & H. J. BUNKER (1927) The Microbiology of Cellulose, Hemicelluloses, Pectin and Gum. London.
TSCHIRCH, A. (1924) Mitt. Naturf. Ges. Bern. **1923,** 19—31.
TSCHIRCH, A. (1925) Handbuch der Pharmakognosie **3,** 2, 965.
VERRALL, A. F. (1939) Phytopathology **29,** 1031—1051.
VIALA, P. (1893) La Maladie de la Vigne, Montpellier, France.
VIALA, P. & G. BOYER (1891) C. R. Acad. Sci. Paris **112,** 1148.
VIALA, P. & G. BOYER (1891) Rev. gen. Bot. **3,** 369—371.
VIALA, P. & G. BOYER (1891) Ann. Ec. Nat. Agric. Montpellier **6,** 152.
VIALA, P. & G. BOYER (1894) C. R. Acad. Sci. Paris. **119,** 61.
VICKLUND, R. E. & M. MANOWITZ (1951) Paint and Varnish Prod. **31,** 2, 8—11, 26.
VIENNOT-BOURGIN, G. (1950) Les Champignons Parasites des Plantes Cultivées, Paris.
WAKEFIELD, E. M. & G. R. BISBY (1941) Trans. Brit. mycol. Soc. **25,** 49—126.
WAKSMAN, S. A. (1916) Soil Sci. **2,** 103—156.
WAKSMAN, S. A. (1917) Soil Sci. **3,** 565—589.
WARCUP, J. H. (1955) Trans. Brit. mycol. Soc. **38,** 298—301.
WARCUP, J. H. (1957) Trans. Brit. mycol. Soc. **40,** 237—262.
WEHMEYER, L. E. (1957) Mycologia **49:** 83—94.
WESTON, W. H. (1949) Q. M. C. Report 60, Microbiology Series Report **15,** 1—76.
WESTERDIJK, J. (1949) Antonie van Leeuwenhoek **15,** 187—189.
WHITE, N. H. (1945) J. Counc. Sci. Ind. Res. Australia **18,** 2, 141—149.
WHITE, W. L. (1945) Letter to E. S. BARGHOORN, 29 Mar. 1945.
WHITE, W. L. (1951) Letter to D. P. ROGERS, 13 Mar. 1951.
WHITE, W. L. et al. (1948) Mycologia **40,** 34—84.
WHITE, W. L., G. R. MANDELS & R. G. H. SIU (1950) Mycologia **42,** 199—223.
WIEGMANN, & A. WOLF (1909) Zbl. Bakt. II, **22,** 657—671.
WIERINGA, K. T. (1953) Atti VI Cong. int. Microbiol. **6(18),** 281—284.
WIERINGA, K. T. (1954) Landbouwkundig Tijdschrift **66,** 292—301.
WIERINGA, K. T. (1955) Z. Pflanzenernährung, Düngung, Bodenkunde **69,** 150—155
WIERINGA, K. T. (1956) Netherlands J. agric. Sci. **4,** 204—209.
WIKEN, T. (1946) Ark. Bot. **33A,** 12, 1—10.
WILSON, M., M. NOBLE & E. G. GRAY (1940a) Nature, London. **145,** 783.
WILSON, M., M. NOBLE & E. G. GRAY (1940b) Nature, London **146,** 492—493.
WILSON, M., M. NOBLE & E. G. GRAY (1945) Trans. Roy. Soc. Edinburgh **61,** II, 12, 327—2340.
WOLF, F. A. & F. T. WOLF (1947) The Fungi, Vol. II, New York.
WYNNE, E. S. & C. L. GOTT (1956) J. gen. Microbiol. **14,** 512—519.

A PROPOSITO DEL LLAMADO MICETOMA MADUROMICOSICO DEL PULMON

Por

FLAVIO L. NIÑO

Laboratorio del Instituto de Clínicas Quirurgicas
Hospital de Clínicas,
Buenos Aires, Argentina

(14.IV.1959)

Desde hace tiempo se conoce la capacidad que poseen algunos mohos de parasitar al parénquima pulmonar o cavidades como bronquiectasias o de otra naturaleza, en las cuales dan el cuadro anátomo-radiológico que se conoce con el nombre de ,,cavidad empastada'', de exteriorización clínica caracterizada por su marcha crónica, hemoptisis a repetición, tos con expectoración hemoptoica y mucopurulenta, con períodos febriles y excelente estado general. La afección parece afectar con preferencia a la mujer y, en cuanto a la localización del proceso, se observa una mayor frecuencia en el lóbulo superior del pulmón izquierdo.

Es lógico pensar que el hongo parásito alcanza su localización pulmonar primitiva o secundaria, por vía aspirativa y que, al encontrar condiciones de medio adecuadas, se multiplique abundantemente y viva al estado parasitario durante mucho tiempo. En éste habitat el parásito adopta la forma y estructura de un grano maduromicósico, de consistencia blanda, coloración pardo-negruzca, forma lobulada y tamaños que oscilan entre 2—3 mm en la observación citada por MACKINNON y col. (1), y 4 ctm. en la de DEVÉ (2).

Desde las primeras observaciones de KOHN (3) y de BOYCE, citado por RÉNON (4), a fines del siglo pasado, en el hombre y en el asno respectivamente, hasta la publicación de MACKINNON y col. mencionada más arriba, pasando por aquellas de MACAIGNE & NICAUD (5), la de KIDBERG, PORAT & NETTER (6) y la de DEVÉ, se habla con más o menos fundamento de la etiología aspergilar de estos procesos en base a un estudio micológico o al hallazgo de cabezas de *Aspergillus* en los cortes histológicos de los granos. Se sindica en la mayoría de los casos a *Aspergillus fumigatus* FRESENIUS, 1850 como la especie en juego.

En el cuadro anátomopatológico y parasitario de ésta micosis, MACAIGNE & NICAUD distinguen aquellas lesiones que se observan en las Aspergilosis pulmonares primitivas con formación de nóddulos granulomatosos fibrosis y alteraciones arteriales que conducen a la trombosis, de aquellas de la Aspergilosis secundaria en las cuales el parásito se desarrolla en alteraciones preexistentes parenquimatosas o bronquiales que facilitan la implantación y multiplicación del hongo, como ocurre en las dilataciones bronquiales y en los quistes aeríferos. En éstas cavidades, la masa miceliana ampliamente entrecruzada y muy abundante, se presenta rodeada por piocitos, células descamadas y por sangre en distintos grados de alteración, constituyendo todo éste conjunto una masa amorfa, pardo-negruzca que ,,empasta'' la luz de la cavidad.

En el Brasil, FLORIANO DE ALMEIDA & CARLOS DA SILVA LACAZ (7), en su trabajo sobre micosis bronco-pulmonares, reproducen en las figs. 20 y 21 granos micóticos de un micetoma pulmonar, posiblemente aspergilar.

En nuestro país, DENIS, HEUDTLASS & MARTI (8), son los primeros en ocuparse del tema al publicar en el año 1943 una observación de megamicetoma pulmonar del lado izquierdo, en una mujer, semejante al caso estudiado por DEVÉ y que los autores consideran como la segunda observación mundial. El diagnóstico de megamicetoma aspergilar se hace en base al cuadro clínico radiológico y al cultivo de *Aspergillus fumigatus* a partir de la expectoración de la enferma.

En 1949, LESTON & MAMÓ (9), estudian una caverna empastada con contenido micótico, comunicando ésta observación a la Soc. de Patología Infecciosa y Tuberculosis del Hospital Muñíz.

BREA, ROGER & GILARDÓN en 1950 (10), publican dos observaciones de quistes pulmonares con contenido micótico en enfermas del Instituto, cuya investigación histopatológica y micológica estuvo a mi cargo; en ambos casos, se trataba de cavidades empastadas del lóbulo superior I.

El estudio de estos casos y de nuevas observaciones, me mueve a escribir el presente trabajo.

MATERIAL Y METODO

El material que sirve de base a éste estudio en sus aspectos histopatológico y micológico, está representado por tres lobectomías practicadas a enfermas que estuviéron internadas en éste Instituto; por una preparación de pieza operatoria obsequiada por el Dr. LATIENDA, procedente de una enferma del Hospital Tornú y por material extraído de una cavidad empastada post-lobectomía, practicada a una enferma por el Dr. M. BREA en una Clínica privada.

Observación No. 1

— R.G. 124.379; Sala I-cama 33 del Inst. de Clín Quir.. Angela C., argentina, de 24 años de edad, soltera, ocupada en quehaceres domésticos.

Antecedentes familiares — Padre fallecido de tumor mediastínico; madre y una hermana sanas.

Antecedentes personales — Sarampión, escarlatina, difteria, rubeola y gastro-enterítis en la infancia. Menarquia a los 11 años, irregular con largos períodos de amenorrea. Apendicítis que no se opera.

Enfermedad actual — Empezó hace 6 años brúscamente de noche, con tos y expectoracón hemoptoica, episódio que se repite a los 15 días. Al mes hemoptisis abundante que duró 6 a 7 días, acompañada de fiebre. Se interna en la Sala VII de éste Hospital, siendo tratada con calcio, vitamina C y sulfatiazol. Alta a los 2 meses en buen estado general, afebril, con escasa tos sin expectoración. La tos y expectoración hemoptoica sólo aparecen durante el período menstrual.

En Noviembre de 1945, nueva hemoptisis, tos y fiebre. Internada en el Hospital Alvear, se le practica neumotórax, penicilina, vitamina C y calcio, mejorando al cabo de un mes.

En Noviembre de 1946, tos y expectoración hemoptoica.

En Marzo de 1947, tos expectoración hemoptoica y fiebre. Se interna en la Sala VII de éste Hospital. La expectoración hemoptoica desaparece y es reemplazada por otra mucopurulenta, numular y escasa; afebril.

Estado actual — Buen estado general con abundante panículo adiposo. Aparato respiratorio: Por delante de la región infraclavicular I., ligera submatitez y rales húmedos, (fig. 1). Resto del organismo nada de particular.

Indice de Katz: 19 (VII-18-47). — Reiterados exámenes de esputos han sido negativos para el bacilo de Koch.

VII-26/947 — En la fecha el Prof. Brea, ayudado por el Prof. Landivar y los Dres. Santas y Martínez, practican una lobectomía superior I.

VIII-13/947 — Alta curada.

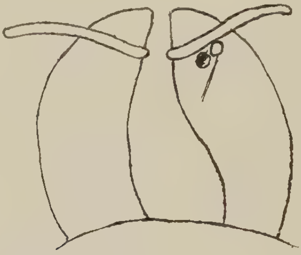


Fig. 1. R.G. 124.379. Esquema mostrando la situación de las cavidades en lóbulo pulmonar superior I.

Pieza operatoria — Lóbulo pulmonar superior I. con superfície pleural despulida y engrosada en su parte anterior y superior. A éste nivel, zona dura del tamaño de una nuez pequeña e inmediatamente por detrás, otra zona de igual volúmen, depresible. Al corte, la primera está representada por una cavidad de paredes lisas y guesas de 3 mm. de espesor, color blanco grisaceo y contenido sólido, de color marrón, adherido a la pared. La segunda formación tiene el mismo aspecto macroscópico, pero su contenido es fluído y mucopurulento. De ésta cavidad parte hacia el hilio un brónquio de 4 mm. de diámetro con paredes engrosadas.

Diagnóstico histopatológico — Inclusión N° 20393 del Lab. del Inst. de Clín. Quir.: Pared quística revestida por epitelio cilíndrico (Dr. Niño).

Inclusón N° 20384 del Lab. del Inst. de Clín. Quir.: El material remitido está representado por conglomerados de micelios fúnguicos rodeados por piocitos (Dr. Niño).

Estudio micológico — La circunstancia de haber fijado la pieza en formol para su estudio histopatológico fué impedimento para practicar cultivos. Con todo, el examen microscópico de cortes realizados con el contenido que empastaba una de las cavidades,

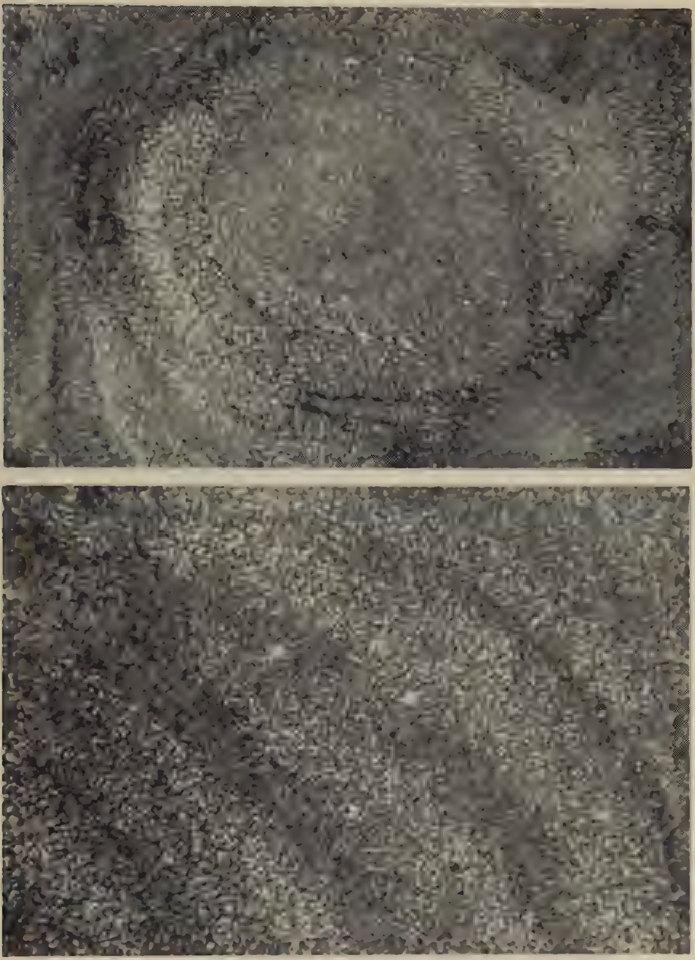


Fig. 2. Aspecto de un corte histológico del ,,grano'' micótico contenido en una de las cavidades de la enferma R.C. 124.379. Muestra el aspecto en franjas o en zonas de la masa miceliana. (Coloración con hematoxilina eosina. Aumento: arriba × 25; abajo × 65).

nos muestra un fieltro miceliano apretado constituyendo masas de hasta 1/2 ctm. de diámetro, con aspecto zonado en franjas concéntricas. (fig. 2). El micelio cortado en distintas insidencias, es pardo-amarillento, más o menos rectilíneo, grueso de 2—4μ de espesor, tabicado, ramificado y con membrana espesa de doble contorno. Se observan dilataciones ampulares intercalares y terminales de 8—10μ de diámetro. No se observan órganos de fructificación ni esporos. Restos de exudados leucocitarios y de detritus necróticos

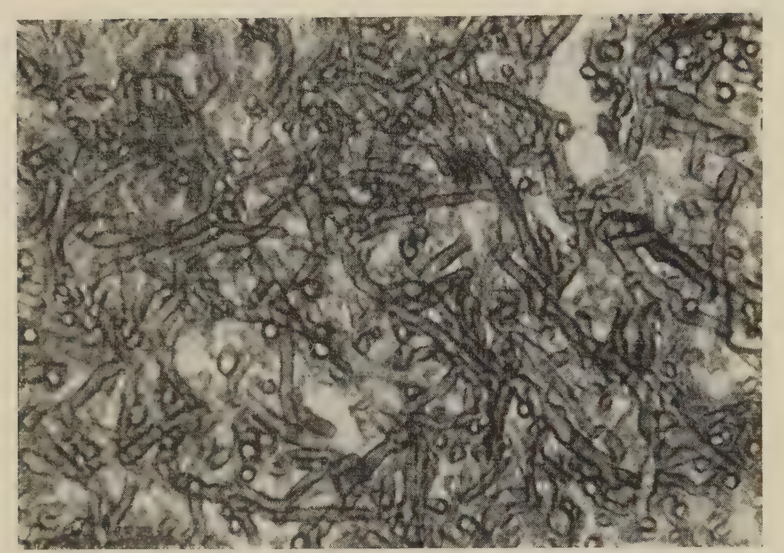


Fig. 3. R.C. 124.379. Un campo de la figura anterior tomado con mayor aumento mostrando la estructura morfológica de la masa miceliana que constituye el ,,grano''. (Aumento × 600).

adhieren a la perifería y aparecen como capa limitante teñidas en un tono más oscuro, (fig. 3).

Observación No. 2

— R.G. 128.953 del Inst. de Clín. Quir., Sala I, cama. Italiana, de 43 años de edad, casada.

Antecedentes hereditarios y personales — Sin importancia.

Enfermedad actual — Empezó hace 1 año y 7 meses con estado gripal febril tos seca y decaimiento, del cual cura en pocos días. Al mes, expectoración hemoptoica que dura 20 días, siendo más abundante por la mañana, afebril. Investigación reiterada de bacilos de Koch, negativa.

En el examen radiológico se aprecia una cavidad de 4 ctm. de diámetro situada en el lóbulo superior D., por debajo de la clavícula, (fig. 4) ocupada por una opacidad que en radiografías anteriores no existía. En las inmediaciones se visualizan otras cavidades más pequeñas.

En Julio de 1948, el Prof. Brea le practica una lobectomía de lóbulo superior D. Alta curada.

Pieza operatoria — Muestra en su vértice una cavidad redondeada del volúmen de una nuez, con paredes lisas, ocupada por una masa de color marrón oscuro, blanduzca, que no adhiere a las paredes.

El material se fija en formol al 20 % y se procede a su inclusión y examen microscópico.

Estudio histopatolócigo y micológico — Inclusión N° 21673 del Lab. del Inst. de Clín. Quir.

Cavidad bronquial sin cartílago, constituída por una pared con infiltrado inflamatorio, espasada en algunos sitios y destruida en otros. El revestimiento está representado por epitelio cilîndrico ciliado. Esta pared se presenta descamada en ciertas zonas y ulcerada en otras. La luz de la cavidad se observa ocupada por una masa miceliana de color pardo-amarillento, constituída por filamentos ramificados, gruesos de 3 y más micrones de espesor, tabicados, con paredes espesas, rectilîneos o más o menos irregulares. Se ven

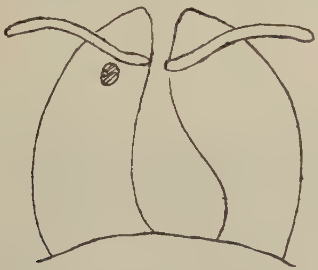


Fig. 4. R.C. 128.953. Esquema mostrando la situación de la cavidad en lóbulo pulmonar superior D.

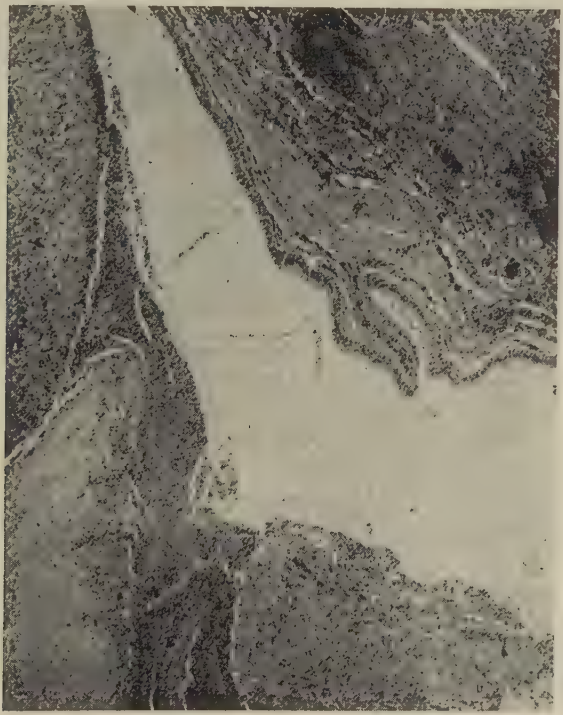


Fig. 5. R.C. 128.953. Fotomicrografía de un corte histológico de la cavidad bronquial y de la masa micótica. (Coloración con hematoxilina eosina. Aumento × 30).

dilataciones ampulares que, hacia la periferia, son más numerosas más irregulares y más voluminosas alcanzando hasta 15μ de diámetro. Estas formaciones semejan clamidosporos intercalares y terminales, (fig. 6). La búsqueda cuidadosa no permite individualizar órganos de fructificación como cabezas aspergilares u otros. En escasa cantidad se observan elementos redondeados de paredes gruesas y 3μ de diámetro que parecen esporos. El conjunto de la masa miceliana recuerda la estructura de los granos maduromicósicos, (fig. 5).

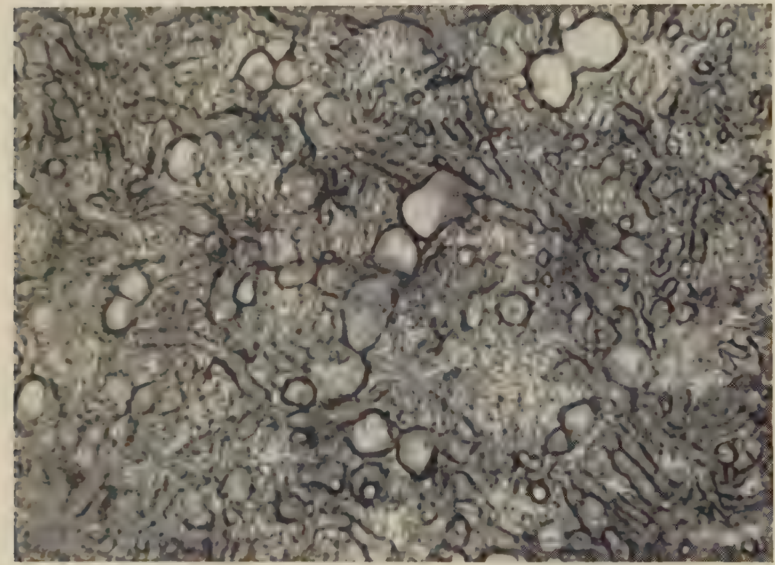


Fig. 6. R.C. 128.953. Fotomicrografía a mayor aumento de un campo microscópico tomado a nivel de la masa miceliana, mostrando la estructura del micelio y de las formaciones ampulares. (Aumento × 600).

Una capa de exudado leucocitario neutrófilo y eosinófilo separa la periferia del grano de la superficie interna de la cavidad. Esta, en la zona ulcerada, es asiento de procesos granulomatosos y necróticos.

Un proceso de bronquioalveolitis exudativa y hemorrágica con atelectasia del parénquima pulmonar, rodea a la formación cavitaria descripta más arriba.

Los vasos sanguíneos son numerosos y no presentan mayores alteraciones.

En síntesis, se trata de una bronquioalveolitis exudativa hemorrágica alrededor de una bronquiectasia ocupada por una masa miceliana que ha ulcerado y necrosado en parte a la pared.

Observación No. 3

— R.G. 175.680 del Inst. de Clín. Quir., Sala I, cama 37. Isolina A. de D, española, de 37 años de edad, casada, ocupada en quehaceres domésticos. Ingresa al Instituto en Junio 6 de 1955.

Antecedentes familiares — Sin importancia.

Antecedentes personales — En 1947 vino a la Argentina. Esposo y 4 hijos, sanos.

Enfermedad actual — Empezó hace 21 años con expectoración hemoptoica en el curso de un período menstrual. El episodio se repite en circunstancias semejantes hasta febrero de 1942 que tuvo una hemoptisis abundante. En los intervalos menstruales se siente perfectamente bien.

Estado actual — El examen radiográfico pulmonar muestra una tumoración de lóbulo superior I. rodeada por un casquete aéreo, (fig. 7). Enferma afebril, en buen estado general. En otros órganos nada de particular. Indice de Katz: 24,5 (VII-8/955).

VI-11/955 — Lobectomía superior I. practicada por el Dr. SANTAS ayudado por los Dres. SPATOLA y RIPETTA.

El post-operatorio se complica con derrame pleural, tromboflebítis de miembros inferiores, expectoración hemoptoica.

En VII-9/955, fallece la enferma con el cuadro de hemorrágia aguda. En la necropsia: Nefrítis crónica congestiva; enfisema de pulmón D., congestión y edema; atelectasia y bronquiectasias en pulmón I.

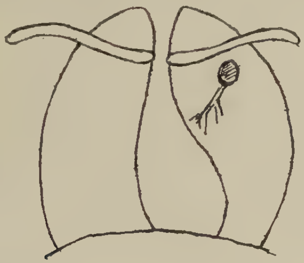


Fig. 7. R.C. 175.680. Esquema mostrando la situación de la cavidad en lóbulo superior de pulmón I.

Pieza operatoria — Quiste empastado de lóbulo superior de pulmón I. del tamaño de una nuez. La cavidad de paredes lisas y engrosadas, se encuentra ocupada por una masa pastosa, de color pardo-negruzco que, en el examen microscópico se muestra constituída en su casi totalidad, por micelio abundantemente entrecruzado, constituyendo un fieltro de elementos rectilíneos o irregulares, tabicados y ramificados, con paredes espesas de 3—4μ de espesor y color pardo-amarillento, (fig. 8). No se observan órganos de fructificación. El conjunto forma pequeños granos redondeados de 2 mm. de diámetro y con aspecto de granos de pimienta.

Parte de éste material se siembra en agar glucosado de Sabouraud y se incuba a temperatura ambiente (16° C.). A las 48 horas, se aprecia el desarrollo de abundantes colónias de un bacilo Gram negativo mucoso, razón por la cual se trata el material con ácido cítrico al 10 y se resiembra en agar glucosado de Sabouraud Los cultivos no desarrollan colónias de hongos a los 37 días de observación.

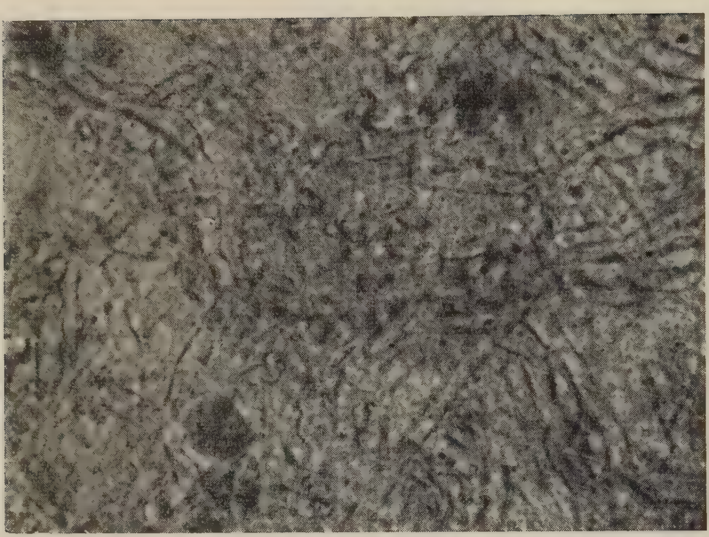


Fig. 8. R.G. 175.680. Aspecto microscópico de la masa miceliana extraída de la cavidad, observada en un preparado fresco entre porta y cubreobjeto. (Aumento × 680).

Observación No. 4

Corte histológico de pieza operatoria rotulada como ,,Micosis pulmonar", (Dr. LATIENDA — Hospital Tornú).

En el examen microscópico se observa un grano micótico voluminoso de contornos policíclicos y estructura radiada, constituído por el entrecruzamiento de micelios gruesos de unos 3 y más micrones de espesor, rectilíneos, con membranas de doble contorno, ramificados y tabicados, de color amarillento. Se observan algunas dilataciones intercalares y terminales en forma de clamidosporos. No se observan órganos de fructificación, (fig. 9). Estos elementos micóticos ocupan una cavidad con revestimiento epitelial o endotelial.

Inmediatamente en contacto con los granos se encuentra una zona de piocitos que, a su vez, contacta con otra de necrosis celular. Por fuera de esta zona de necrosis que puede contener algunos micelios o trozos de micelios sueltos, se observa un proceso inflamatorio crónico diseminado, escleroso y denso en cuyo seno se ven gruesos y numerosos vasos sanguíneos.

A distancia de este foco principal, se observan otros más pequeños, al parecer más recientes, constituyendo pequeños granulomas con escasas células gigantes del tipo Langhans, alrededor de conglomerados micelianos. En la perifería de la zona de esclerosis se ven sufusiones sanguíneas en napa y luego una zona de atelectasia pulmonar con alveolos y bronquiolos aplastados, (fig. 10).

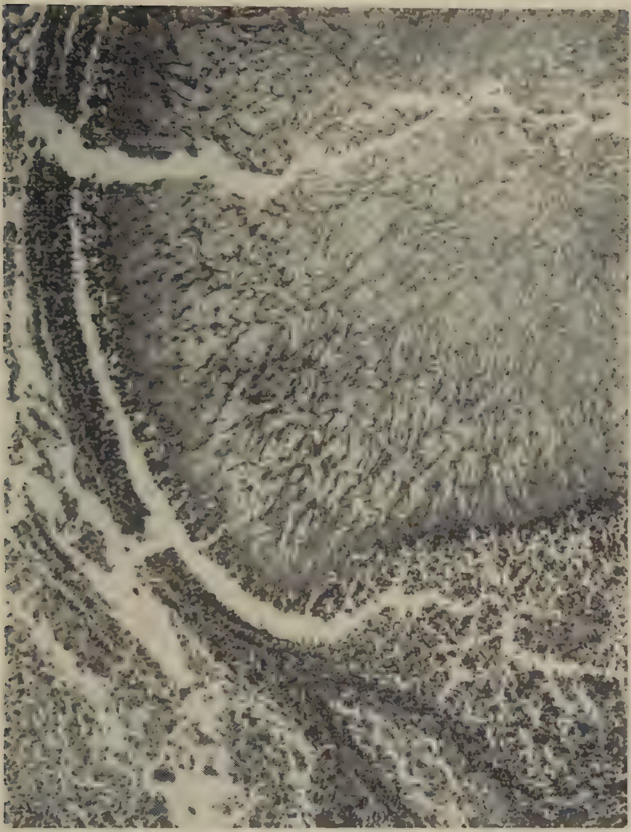


Fig. 9. Observación N° 4. Corte histológico de la cavidad bronquial y del voluminoso grano micótico que ocupa dicha cavidad en una visión con poco aumento. (Coloración con hematoxilina eosina. Aumento × 80).

En síntesis, se está en presencia de una maduromicosis en cavidad pulmonar con intensa fibro-esclerosis y presencia de granulaciones micóticas a distancia.

Observación No. 5

En Mayo 12 de 1957, se recibe enviado por el Prof. Brea desde una Clínica privada, un tubo estéril conteniendo el producto extraído de una cavidad quística situada en lóbulo pulmonar I., de la enferma Isabel de A. El pedido de estudio micológico, se acompañaba del esquema que ilustra la fig. 12.

Estudio micológico — El material está representado por una masa fragmentada de color pardo-negruzco, de consistencia blanda que, en preparados frescos entre porta y cubreobjeto, muestra estar constituída en el examen microscópico por un fieltro de abundantes

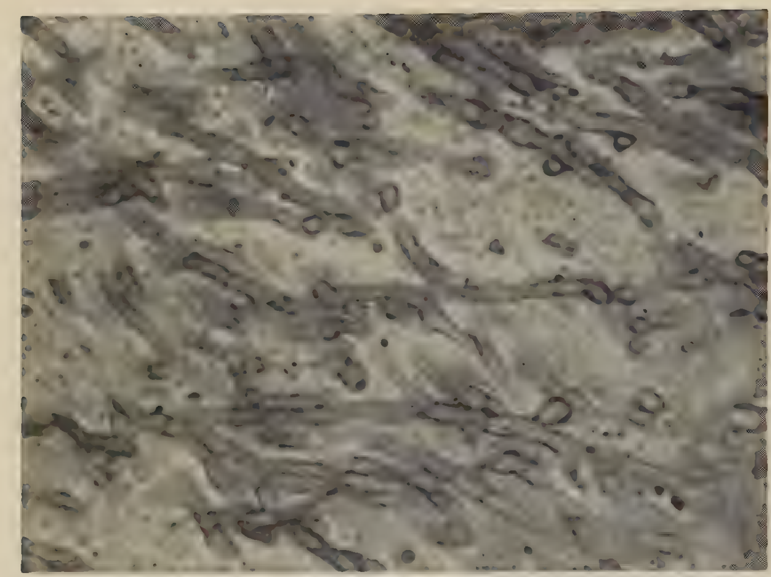


Fig. 10. Fotomicrografía de un campo de la figura anterior a nivel de la masa miceliana tomada con mayor aumento. Las formaciones que semejan esporos no son tales, se trata de filamentos seccionados transversalmente. (Aumento × 600).

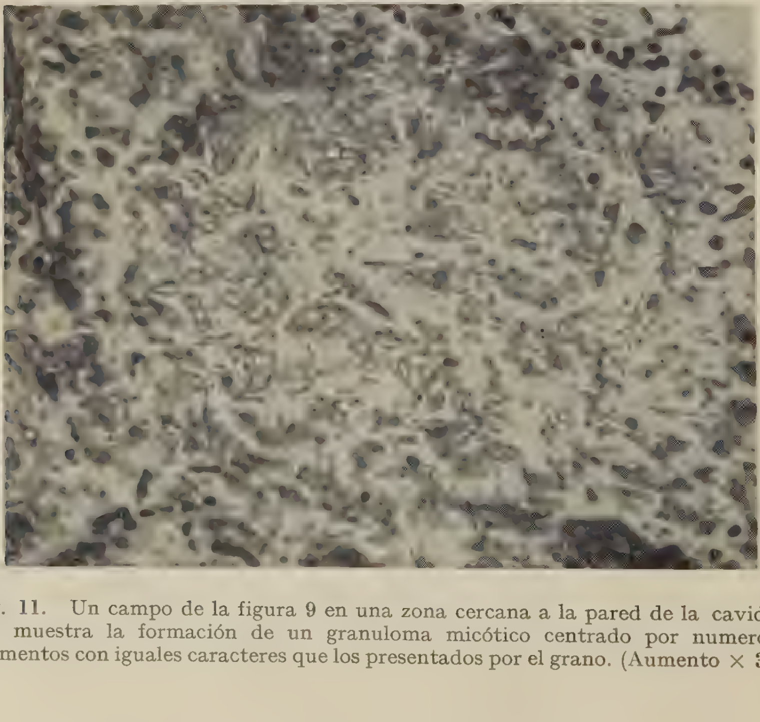


Fig. 11. Un campo de la figura 9 en una zona cercana a la pared de la cavidad; nos muestra la formación de un granuloma micótico centrado por numerosos filamentos con iguales caracteres que los presentados por el grano. (Aumento × 360)

micelios de color amarillento, rectilíneos o más o menos irregulares en sus trayectos, tabicados, ramificados, de 3—4μ de espesor, provistos de membrana gruesa y presentando dilataciones intercalares y terminales como clamidosporos, de 10—12μ de diámetro, (fig. 13). No se observan órganos de fructificación.

Parte de éste material se siembra en agar glucosado de Sabouraud y el resto se fija en formol al 20 % para ser incluído en parafina.

Aspecto microscópico de los cortes — El conglomerado miceliano forma granos redondeados con una estructura zonada perfectamente delimitada por una coloración rosada de la eosina a ese nivel, (fig. 14) que lo hace en un tono más subido. Estas franjas estan ocupadas por gran cantidad de formaciones ampulares esféricas o fusiformes que se interpretan como clamidosporos, (fig. 14). El resto de los granos estan representados por filamentos con la morfología señalada más arriba, cortados en distintas insidencias, (fig. 15). También en ésta oportunidad, no fuéron observados órganos de fructificación ni esporos característicos.

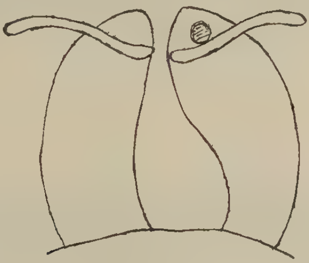


Fig. 12. Esquema de la situación ocupada en el lóbulo superior del pulmón I por la lesión de la enferma Isabel de A.

Cultivos:

Aspecto macromorfològico — A los 10 días se observan colónias esféricas, secas, de color blanco-ceniza, muy adherentes al médio, cubiertas por hifas aéreas cortas; de 2—3 mm. de diámetro; dorso no pigmentado.

A los 23 días, las colónias miden alrededor de 1/2 ctm. de diámetro; la parte central se ha hundido con aparición de gruesos pliegues. en el ápice de algunas colónias aparece una coloración verdosa ténue. La colónia es dura y muy adherente al medio; éste se deprime alrededor de la misma. El dorso presenta una coloración ligeramente rosada.

A los 40 días las colónias tienen 2 ctm. de diámetro, es más o menos aplanada y con superficie ampliamente plegada formando surcos y pliegues muy irregulares; el color no ha variado mayormente como tampoco el aspecto seco y su gran adherencia al medio de cultivo. En algunas zonas de la perifería se observan porciones en las cuales las hifas aéreas son más largas y con un color blanco de nieve, (fig. 16).

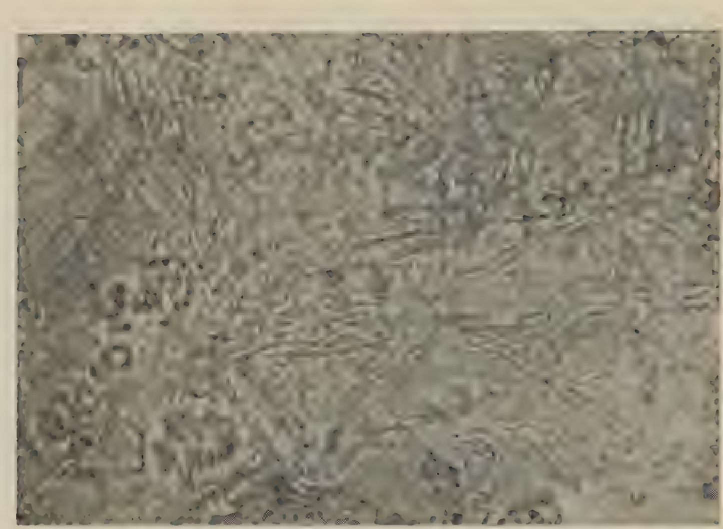


Fig. 13. Aspecto de la masa miceliana extraída de la cavidad bronquial de la enferma Isabel de A., en observación microscópica de un preparado fresco entre porta y cubreobjeto. (Aumento × 600).

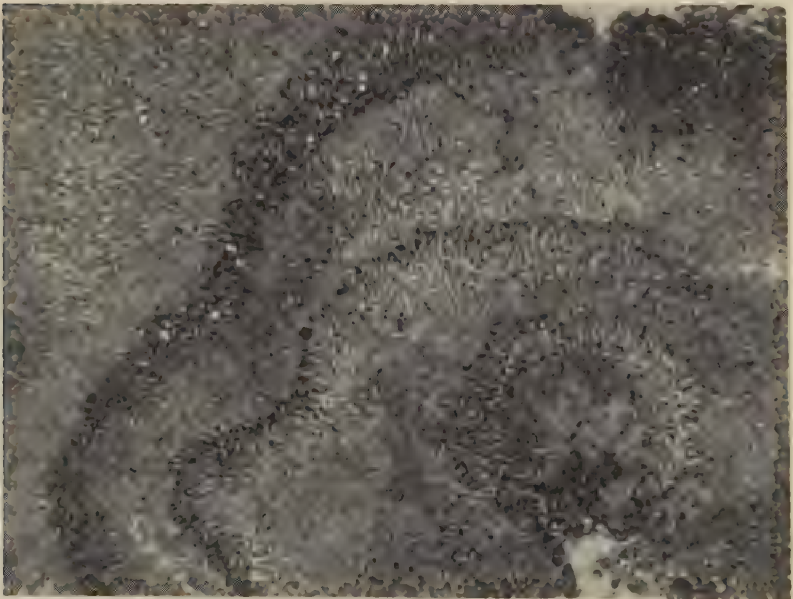


Fig. 14. Fotomicrografía de un corte histológico de la masa miceliana extraída de la cavidad bronquial de la enferma Isabel de A.; muestra el aspecto zonado que adquiere la formación micótica (coloración con hematoxilina eosina. Aumento × 80)

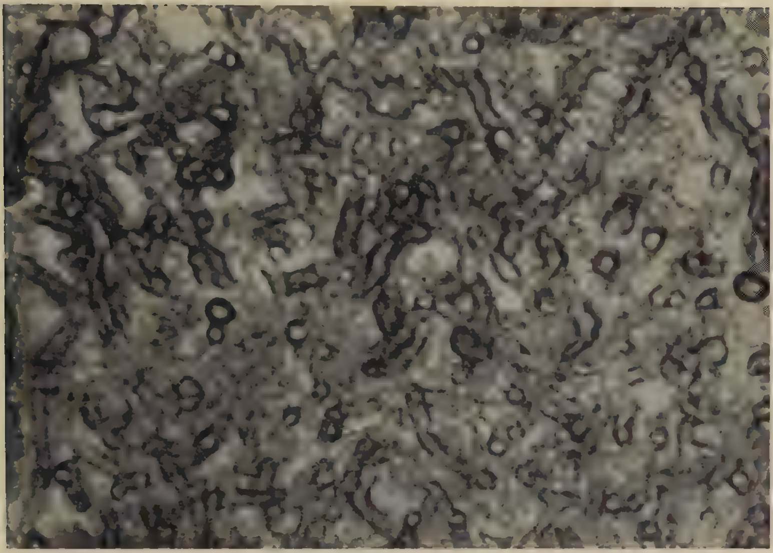


Fig. 15. Fotomicrografía de un campo de la figura anterior tomada con mayor aumento para mostrar el aspecto de los filamentos en la masa miceliana. (Aumento × 600).

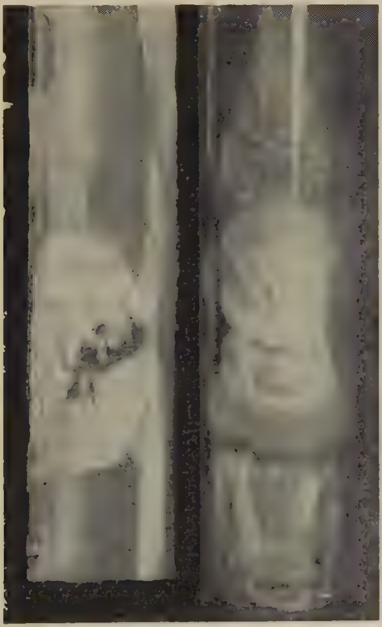


Fig. 16. Colónias del hongo aislado de la masa miceliana extraída de cavidad bronquial a la enferma Isabel de A. A la izquierda, colónia sobre Sabouraud glucosado de 40 días de edad. A la derecha, colónia shore agar mosto de 20 días de edad. (Tamaño natural).

En agar-mosto, el crecimiento es exuberante y rápido. Las colónias son de color blanco opaco, secas y con pliegues radiados a partir del centro elevado.

En papa y zanahoria glicerinada al 5 %, hay poco desarrollo en los primeros días para hacerse más abundante en los siguientes, en especial sobre zanahoria. Las colónias son de color blanco y con aspecto seco.

En agar-tierra, el crecimiento es mediocre.

En pasta de papel, hay escaso desarrollo. No se aprecia acción sobre la celulosa.

En placas con agar-almidón, hay buen desarrollo. No se aprecia acción amilolítica.

En gelatina por punción y en pico de flauta, incubada a 16° C., el desarrollo es lento y no se observa licuación del medio a los 60 días de observación.

La investigación de hemolisinas, bacteriolisinas y de la acción bacteriostática, ha dado resultado negativo.

En suero coagulado y glucosado, crece licuando al medio.

ACCION SOBRE LOS ALCOHOLES: (VIII-10/957

	Cultivos a 37° C.			
	7 días	15 días	21 días	30 días
Alcohol etílico	+	+	+	+
Alcohol metílico	++	++	+++	+++
Alcohol amílico	—	—	—	—
Testigo	—	+	+	+

Desde el comienzo de la observación se aprecia un buen aprovechamiento del alcohol metílico.

ACCION SOBRE LOS HIDRATOS DE CARBONO: VIII-10-57

	Cultivos a 37° C.			
	7 días	15 días	21 días	30 días
Glucosa	+	+++	+++	+++
Maltosa	++	++++	++++	++++
Sacarosa	+	+++	+++	+++
Galactosa	—	++	+++	+++
Lactosa	—	++	++	++
Testigo	—	+	+	+

Se aprecia el aprovechamiento de las distintas fuentes ensayadas. Desde el comienzo de la observación ha sido la maltosa la que ha permitido un crecimiento más rápido y exuberante.

ACCION SOBRE EL NITROGENO: VIII-10/957

	Cultivos a 37° C.		
	7 días	15 días	21 días
Nitrato de potasio	++	+++	+++
Urea	+	+++	+++
Asparragina	++	++++	++++
Peptona	+++	++++	++++
Sulfato de amonio	++	++	++
Testigo	+	++	++

Si bien ésta experiencia muestra que no es menester la presencia de substancias nitrogenadas para obtener un buen desarrollo del hongo en estudio, nos enseña que las distintas fuentes ensayadas benefician su crecimiento, siendo particularmente aptas la asparragina y la peptona.

ACCION PROTEOLITICA: VIII-10/957

	Cultivos a 37° y 16° C.		
	7 días	15 días	21 días
Suero coagulado	+	+	+
Albúmina de huevo	—	—	—
Gelatina	—	—	—
Leche	+	+	+

Acción proteolítica muy debil sobre el suero coagulado y la leche.

Aspecto micromorfológico — Micelio grueso, más o menos flexuoso, de 3—4μ de espesor, más o menos hialino, con paredes gruesas y membrana que ofrece un doble contorno, irregularmente ramificado y tabicado. En la parte central de la colónia el micelio presenta un contorno más regular, tiene coloración amarillenta y se parece al aspecto observado en los productos del organismo, (fig. 17-1).

En cultivos de 23 días de edad, el micelio es hialino y sumamente irregular, tabicado, en parte moniliforme y presenta a los costados una serie de formaciones mamelonadas y angulosas que confieren al conjunto un aspecto que se caracteriza por la falta de regularidad. El contenido del micelio es algo granuloso. Se observa la formación de algunas dilataciones ampulares en el extremo de ciertas hifas, (fig. 17-2).

A los 40 días, el aspecto micromorfológico no ha cambiado mayormente y es comparable al señalado con anterioridad. Se observa una mayor irregularidad en la morfología de las hifas que muestran ensanchamientos en su trayecto y un número más grande de dilataciones terminales, algunas voluminosas, hialinas y con contenido granuloso refringente. Continúa como micelio estéril, sin esporos y sin órganos de fructificación, (fig. 17-3).

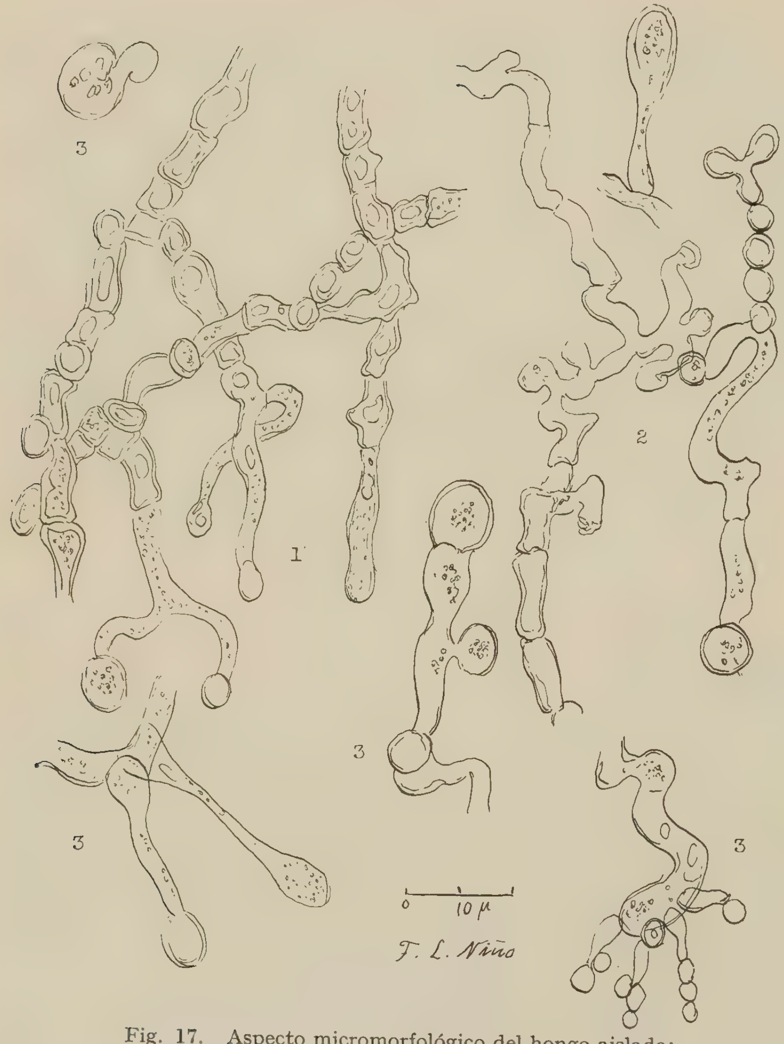


Fig. 17. Aspecto micromorfológico del hongo aislado:
1) En cultivos de 10 días edad,
2) En cultivos de 23 días de edad,
3) En cultivos de 40 días de edad.
(Imágenes dibujadas con auxilio de la cámara clara).

En cultivos sobre el medio de Czapek a 37° C. de 11 días de edad, el micelio conserva en líneas generales, el caracter irregular señalado más arriba; pero, en ésta oportunidad se puede observar el desarrollo de órganos de fructificación en el extremo de conidióforos, sumamente anómalos en su morfología, que semejan cabezas aspergilares con una corona de fiálidas simples, cortas y largas igualmente irregulares, como puede apreciarse en la fig. 18-1). Los conidióforos

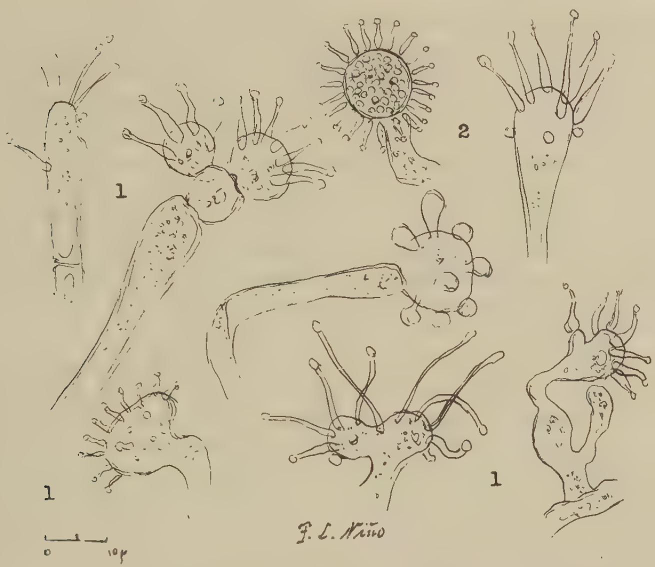


Fig. 18. Organos de fructificación observados en cultivos sobre el medio de Cazapek:
1) Cabezas aspergilares anómalas,
2) Cabezas aspergilares normales.
(Imágenes dibujadas con auxilio de la cámara clara).

son en algunos casos cortos y robustos, de unos 50μ de longitud por 6—8μ de ancho y, en otros, largos, más regulares, algo flexuosos, con más de 100μ de longitud por 4—5μ de ancho. Estos últimos se ensanchan progresivamente hasta terminar en una cabeza irregular y anómala, que son las más, o bien rematan en una dilatación esférica de 10—12μ de diámetro, de contorno regular, sobre cuyo ápice se implanta una serie de fiálidas cortas y regulares de 5—8μ de largo que confieren al conjunto el aspecto de típicas cabezas de *Aspergillus*. Estas formaciones son raras de observar, (fig. 18-2). No se ven esporos.

DISCUSION

El estudio de nuestras observaciones y de aquellas citadas en la bibliografía, nos enseña que, en la mayoría de los casos, la masa micótica estaba alojada en cavidades bronquiectásicas que seguramente ya existían antes del arribo y colonización del hongo. En éste sentido comparto la opinión de BREA y col. al negar al hongo parásito un papel preponderante en la patogenia de la dilatación bronquial como factor primitivo de semejante alteración. De acuerdo a lo observado, el proceso se desarrollaría de la siguiente manera: El hongo es aspirado hasta una cavidad preexistente donde encuentra condiciones favorables para su desarrollo y colonización. Pregresivamente el crecimiento miceliano aumenta de volúmen hasta formar una masa compacta que, conjuntamente con la acumulación de secreciones y coágulos sanguíneos empastan la cavidad y bloquean el orificio de drenaje, sufriendo las paredes de la misma, la acción expansiva que le imprime progresivamente el contínuo crecimiento y multiplicación del hongo que puede llegar a constituir masas con volúmenes considerables como el descripto por DEVÉ en su ,,Megamicetoma''.

En la observación comentada por MACKINNON y col., también el grano micótico se habría desarrollado en cavidades bronquiales según opinión del patólogo que realizó el estudio histopatológico; es decir, semejante en sus aspectos histo-micológicos a los que hemos estudiado más arriba.

Si bien el aspecto microscópico que presenta la masa miceliana en los cortes resulta muy semejante a la que ofrecen los granos que se observan en los verdaderos micetomas maduromicósicos, no creo sea ventajoso aplicar para los casos que estudiamos el mismo concepto patogénico que en aquellos. El micetoma maduromicósico es una micosis verdadera y primitiva que nace y se desarrolla como consecuencia de la penetración de uno de sus agentes en el seno de un tejido sano; la observación de PARINI, RIBOTTO & SPROVIERI (11), recientemente descripta, es bien demostrativa a éste respecto. En los casos que estudiamos, el hongo ha colonizado en una lesión preexistente a la cual complica por una acción especialmente mecánica. Por ello, preferimos hablar de cavidades con contenido micótico o maduromicótico y no de micetoma pulmonar maduromicótico.

En lo que respecta al moho que parasita estas cavidades pulmonares, ya hemos dicho que la mayoría de los autores lo identifican con el *Aspergillus fumigatus*, unos con fundamento y otros, sin mayor apoyo micológico. No basta la comprobación del hongo en la expectoración ya que, en nuestro medio, al estudiar la flora micológica en exámenes rutinarios de esputos, hemos encontrado a especies del Género *Aspergillus* en el 14,64 % de los casos en sujetos que no tenian el cuadro clínico-radiológico que acompaña al proceso que estudiamos, (12). Sólo en pocos casos se han podido efectuar cultivos a partir del contenido de las cavidades, cuando el cirujano, ya advertido, proporciona al laboratorista un material recogido en condiciones óptimas, sin agregado de formol.

De las 5 observaciones que han sido estudiadas más arriba, en 3 el material fué fijado en formol para exámenes histopatológicos haciendo imposible toda tentativa de cultivo; en las otras 2, el material recien extraído de las cavidades fué sembrado en medios apropiados, fracasando uno de ellos, no así el otro que nos ha permitido aislar un hongo y realizar el estudio micológico del mismo.

Si nos referimos al aspecto microscópico de los ,,granos'' o masa miceliana examinada en preparados fresco entre porta y cubreobjeto y en los cortes histológicos, nos sorprende la uniformidad de los caracteres morfológicos en todos los casos estudiados, como asimismo, la ausencia de cabezas aspergilares u otros órganos de fructificación, pese a numerosos cortes practicados. Estos resultados negativos me llevaron a dudar en un princípio de la etiologia aspergilar en los casos estudiados y a suponer que bien pudiéra ser otra la especie con capacidad de colonizar y empastar las cavidades bronquiales.

En el único que se logró cultivar al agente parasitario, el estudio macro y micromorfológico del mismo no ha permitido caracterizar a especies del Género *Aspergillus* en cultivos de más de un mes de edad y, ni siquiera el examen microscópico de cortes histológicos de una colónia bien desarrollada, ha sido capaz de suministrar algún punto de apoyo para aceptar ésta u otra etiologia, ya que, en todas las circunstancias el hongo estudiado se mostraba como *sterile micelia.* Unicamente los repiques sobre el medio de Czapek mostráron el desarrollo de cabezas aspergilares sumamente atípicas, circunstancia que nos permitió situar a esta especie dentro del Género *Aspergillus* MICHELII. Esto dicho, viene a corroborar las dificultades que se presentan para suponer la existencia de una etiología determinada cuando no se cuenta con un estudio micológico lo suficientemente completo como para caracterizar un Género y más aún, una especie.

CONCLUSIONES

Se efectúa el estudio de 5 observaciones del llamado ,,Micetoma maduromicósico de pulmón'' en sus aspectos, histopatológico, micológico y clínico.

Todas ellas pertenecen a mujeres y configuraron el cuadro de cavidad bronquial ,,empastada'', anotando el predominio de su localización en el lóbulo superior izquierdo.

Se señala la uniformidad de los caracteres morfológicos que presenta la masa miceliana llamada ,,grano'' en todos los casos estudiados, en los cuales no fué posible individualizar la existencia de órganos de fructificación que permitiéran una clasificación, cuando más no fuera, genérica del hongo observado.

Se critica la aplicación del término Micetoma para éstos casos con igual criterio que el clásico, que supone una enfermedad micótica primitiva, razón por la cual se prefiere hablar de cavidad con contenido micótico o maduromicótico.

En el único caso que se logró cultivar al hongo parásito, el estudio micológico del mismo permitió aislar una especie del Género *Aspergillua* con caracteres morfológicos sumamente atípicos.

SUMMARY

The author presents the study of five observations of the so-called ,,Maduromycosis-mycetoma of the lungs'' in their clinical, histopathologic and mycologic aspects.

The pathologic features in all these cases have been found in bronchial cavities of women, situated in the upper lobe of the lung. A compact mycelial mass, the ,,grain'', filled up these cavities.

In one of the five cases a fungus was cultivated which was classified as belonging to the Genus *Aspergillus* Michelii, with abnormal and atypical features.

Short criticism is presented about the concept ,,Maduromycosis mycetoma of the lung'', applied by authors designating this process.

Bibliografia

1. Mackinnon, J. E., Ferrada, L. V. & Montemayor, L., Investigaciones sobre las Maduromicosis y sus agentes. Anal. Facultad de Med., **34,** (1-2-3): 231—309. Montevideo, 1949.
2. Devé, F., Une nouvelle forme anatomo-radiologique de mycose pulmonaire primitive. Le mégamycetome intra-bronchectasique. Arch. Méd. Chir. de l'App. Resp., **13** (5): 337—361. 1938.
3. Kohn, H., Ein Fall von Pneumomycosis Aspergillina. Dtsch. med. Wschr., **19:** 1332—1333. 1893.
4. Rénon, L., Etude sur l'Aspergillose chez les animaux et chez l'homme. Masson et Cie., Ed., Paris, 1897.
5. Macaigne & Nicaud, Aspergillose primitive du poumon avec artérite pulmonaire obliterante. Bull. Mém. Soc. Méd. des Hôp. Paris, 1920.
6. Kidberg, M. L., Parat, M. & Petter, H., Tumeur mycosique du poumon (Aspergillose pulmonaire primitive pseudo-cancéreuse). Presse Méd., 1936, N° 92.
7. de Almeida, F. P. & da Silva Lacaz, C., Micoses Bronco-Pulmonares. Comp. Melhoramentos de São Paulo. São Paulo, 1942.
8. Denis, R., Heudtlass, A. P. & Marti, J. A., Micosis pulmonar de forma tumoral: Megamicetoma. La Prensa Méd. Argent., **30,** (24): 1189—1199. 1943.
9. Leston, J. M. & Mamó, H. R., Caverna empastada de contenido micótico. Soc. de Patol. infecc. y Tuberc. del Hosp. Muñíz. (Reunión del 24 de Nov. de 1949).
10. Brea, M. M., Roger, V. N. & Gilardón, A., Quistes pulmonares con contenido micótico. Rev. Acos. Méd. Argent., **64:** 207—210. 1950.
11. Parini, F., Ribotto, L. & Sprovieri, L., Pneumomicosi Cronica (Aspergillosi?) con generalizzazione pluriviscerale. Mycopath. Mycol. Appl. 8 (1): 27—46. 1957
12. Niño, F. L. & Nolte, A. I., Estudio de la flora micológica de la expectoración, en la Ciudad de Buenos Aires. La Prensa Méd. Argent., **40** (20): 1212—1215. 1953.

EXPERIMENTAL FUNGUS GRANULOMA AS SEEN FROM COMPARATIVE EXANTHEMATOLOGY [1]

KASUKE ITO, M. D.

Department of Dermatology,
Gifu Medical College, Gifu-City, Japan.

(15.VI.1959)

For years, our interest has been directed to fungus immunobiologic eruptions (inclusive of "id" skin manifestations) artificially produced by fungus reinfection. A series of investigations in this direction is motivated not only by a desire for a better understanding of fungus skin reactions ("id" in particular), but also by the desire to produce information on the comparative exanthematology of these skin reactions with similar reactions due to other etiologies (tuberculid, syphilid etc., in particular). Guided by this idea, previously the author and his collaborators performed experimental study on some tr. (mentagr., var. ast.) skin reactions, such as corymbiform eruption, erythema indurativum like phlebitis, arteritis and a variety of tuberculoid granulomas, etc., using rabbits as test-animals. In this paper experimental fungus granuloma is presented as seen from comparative exanthematology, based on our further personal experience (experimental and clinical data recently obtained by the author and his collaborators). Without going into technical minutes, main findings obtained are summed up under the following five items.

I. **Fungus Cutaneous granuloma**

This is experimental production of fungus granulomas on the ears of male rabbits due to a second dorsal inoculation with tr. mentagr., var. ast. Experimental method(Tr. mentagr., var. ast. was cultured on Sabouraud's ager for 4 weeks. For this the fungus was freshly isolated from inoculated rabbits. Adult albino rabbits were infected with the tr.-suspension according to BLOCH's method. 1 month after the "healing" of the typical trichophytia on the back skin, reinoculation was made in the same manner as above given. Result: Erythematous nodules on the ears (Fig. 1) manifested themselves

---

[1] Read before the sixth International Congress on Tropical Medicine and Malaria, Division A Section: 3 Sub-section: 4-Mycotic Infections —, Lisbon, 5—13 September, 1958.

1—2 weeks after the 2nd inoculation under certain experimental condition. The nodules followed the quick reaction of the reinoculation lesion on the dorsum, whereas corymbiform lesions were occasionally produced after the 2nd inoculation with the tr. Histologically, the nodules, active stage, showed tuberculoid structure centering around central necrotizing zone (containing fungi-elements as shown by staining-method, Fig. 2, Fig. 3). This tuberculoid granulomas were preceded or accompanied by fungus phlebitis (Fig. 6) and occasionally arteritis (Fig. 5) on the ears.

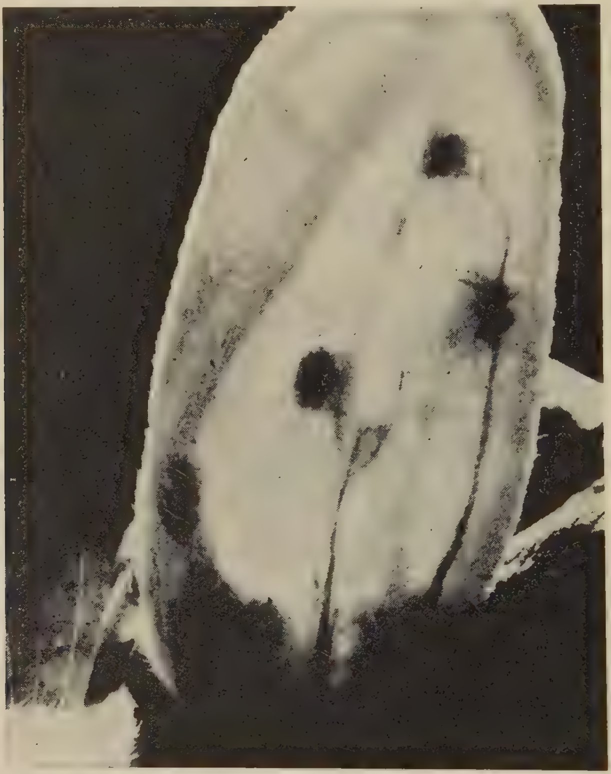


Fig. 1. Showing dermal erythematous indurated nodules on the ear of the rabbit, after the second dorsal inoculation with tr. mentagr. var. ast.

These skin reactions (granuloma, phlebitis and arteritis) are comparable to cases of human fungus cutaneous reaction and skin tuberculosis in man. Besides, fungus perifolliculitis granulomatosa was produced. The histology of the perifolliculitis (Fig. 4) closely resembled that of nodular granulomatous perifolliculitis caused by trichophyton in man and that of lichen trichophyticus. Also, it is noted from the point of view of comparative exanthematology that

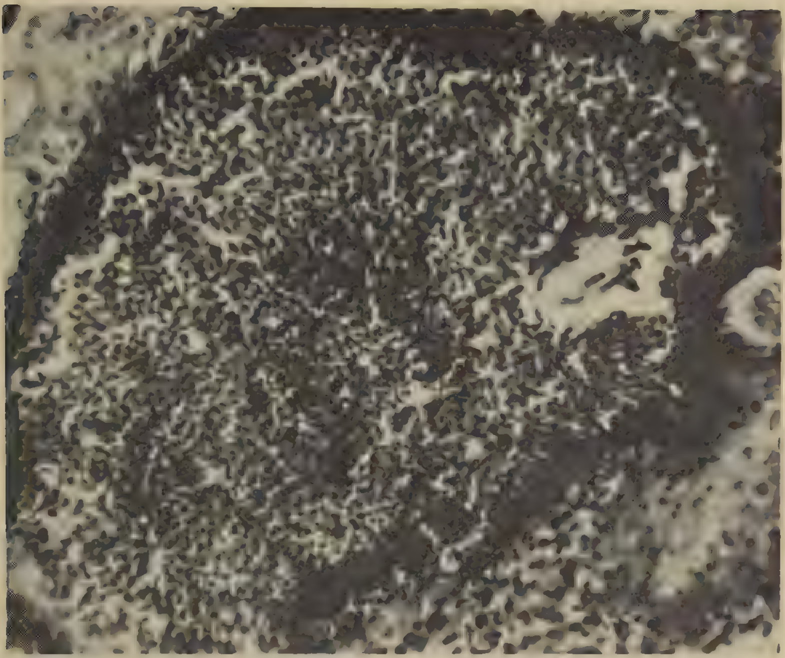


Fig. 2. Showing the histology of fungus abscess containing fungielements in the dermis of the ear of the rabbit 20 days after the onset of the erythematous nodules on the ear. This fungus abscess was surrounded by the granulomatous infiltrate.

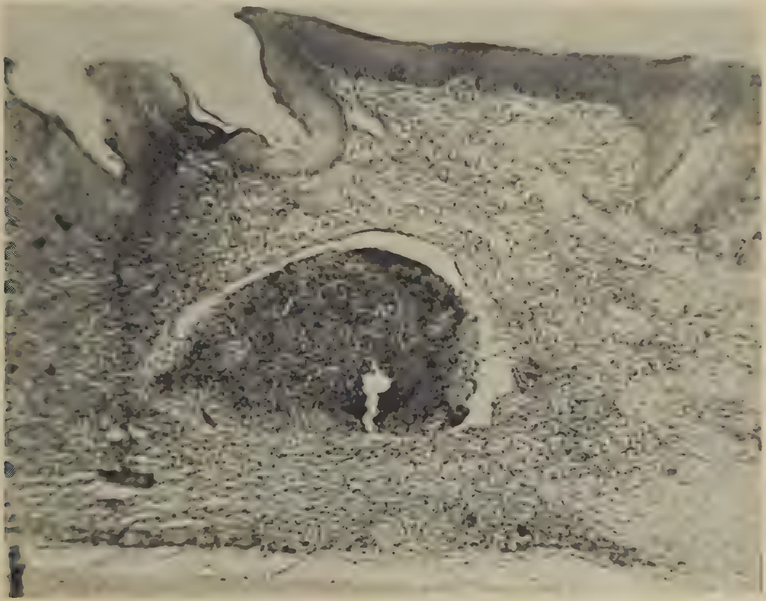


Fig. 3. Showing epithelioid cell response surrounding a large abscess (containing fungielements). This granulomatous infiltrate appeared 1 month after the onset of the erythematous nodules on the ear of the rabbit.

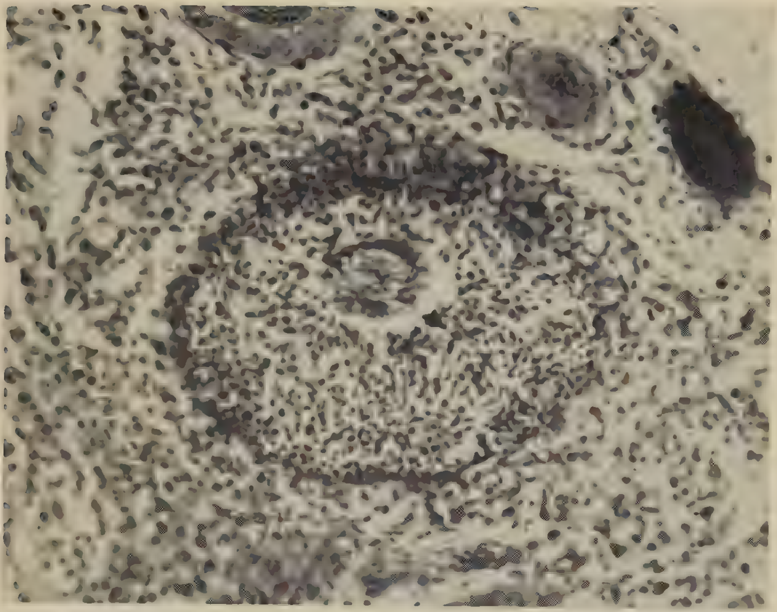


Fig. 4. Showing histologic feature of fungus perifolliculitis granulomatosa on the ears of the rabbit 10 days after the 2nd dorsal infection with tr. mentagr., var. ast.

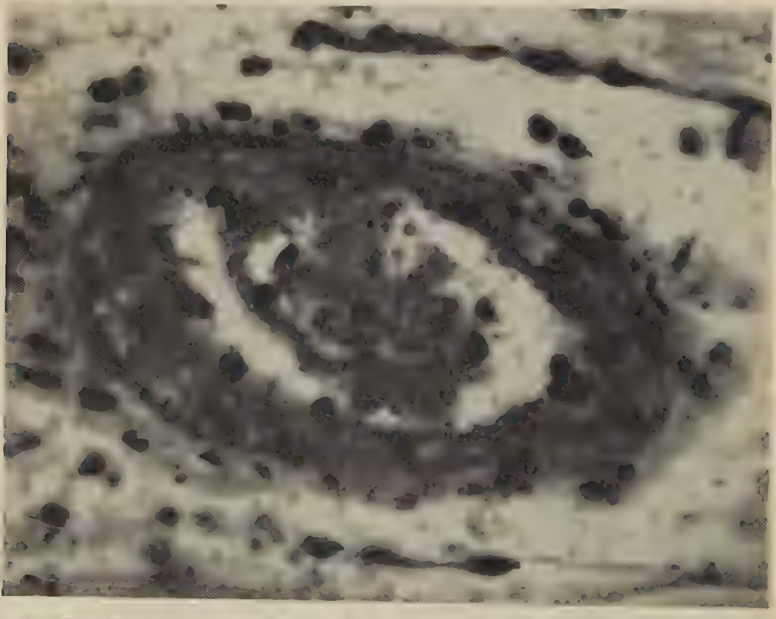


Fig. 5. Showing the histology of arteritis characterized by hyaline thrombus, fibrinoid degeneration of vessel wall, and the intensive perivascular edema (20 days after the onset of the fungus "id" eruption on the ear of the rabbit) caused by fungus dorsal inoculation.

we observed, in cases of lupus miliaris disseminatus faciei (tuberculin skin reaction negative), similar histologic feature as above given (The feature showed peri- and para-folliculitis granulomatosa lacking tuberculous caseation).

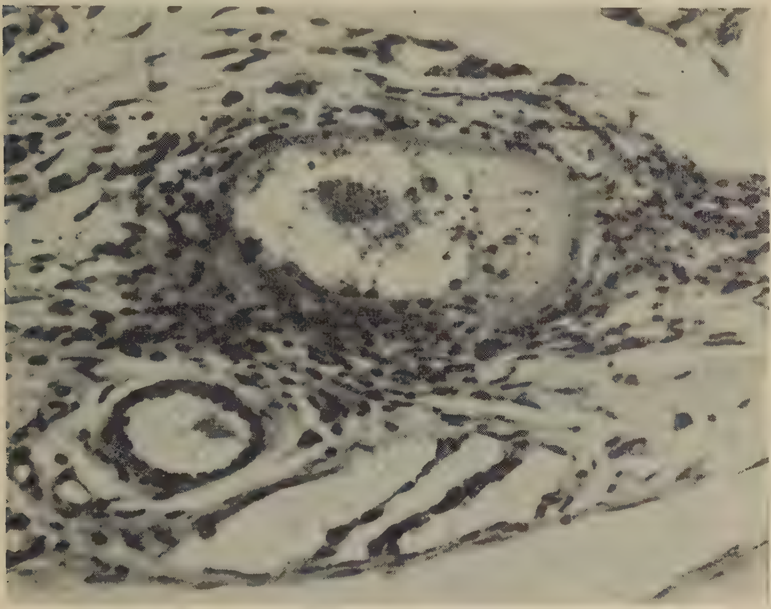


Fig. 6. Showing the histology of phlebitis, characterized by a hyaline-like thrombus and a severe perivascular infiltrates which consisted of monocytes, lymphocytes and plasma cells ("id" on the ear of the rabbit caused by tr. mentagr., var. ast. dorsal reinfection).

II. Hematogenous disseminate reinfection with the tr.

Similar fungus granuloma, perifolliculitis granulomatosa, fungus thrombophlebitis and vascular granuloma as in I given were also produced on the ears of rabbits by reinfection with the tr. ast. via cardiac route. The rabbits had previously had a first dorsal inoculation with the tr. Attention is called to characteristic feature of the thrombus occulded in the damaged venous lumen: Frequently, the thrombus consisted of rosette-like aggregates of platelets and leucocytes. Besides, before the said cardiac reinfection, blood was collected by cardiac puncture, then the blood thus obtained was heparinized, and the heparinized blood was mixed with the tr. ast. suspension, then placed at 37° C for 30 minutes, and smears were drawn in usual manner. Thus, frequently similar rosette-like accumulation of platelets and leucocytes was also produced in vitro.

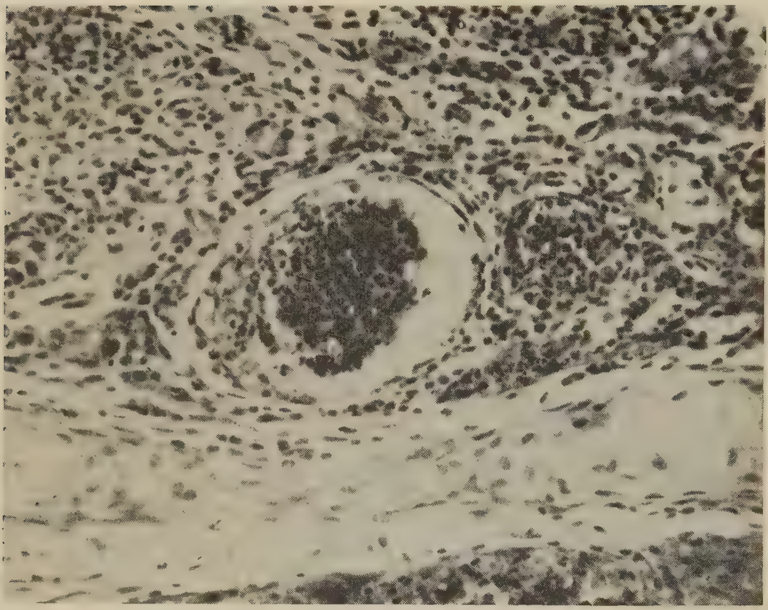


Fig. 7. Showing intensive thrombus, damaged venous vessel wall and exsudative inflammatory infiltration which effects the venous wall and its surrounding (2 days after the onset of the erythematous nodules on the ears of tr. ast. immune rabbits due to the intracardiac reinfection with the tr.-suspension in the rabbit). This animal had previously had the first dorsal infection with the tr.

III. Lung feature of trichophyton granuloma

Just as experimental cutaneous granuloma due to the tr. is of interest from comparative exanthematology of mycosis with tuberculosis, fungus reactions produced in the visceral organs attracts our attention from the same view-point. 1 week after the "healing" of a first inoculation with the tr. ast. on the dorsum of the rabbit, the tr. suspension was injected via venous route. 2—3 weeks after the intravenous infection, test animals were destroyed. The present attention is called to fungus lesions at the site of lung-parenchym: Conspicuous in both fungus granuloma. Histologically, this lung-granuloma shows tuberculoid structure with or without central necrotizing area, mingled with giant cells (foreign-body type and Langhans-type); The necrotizing portion consisted of disintegrated fungi-elements and necrosis of polymorphs with nuclear debris; in the giant cell of Langhans' type, disintegrated fungielements, nuclear debris of polymorphs and lymphocytes were phagocytized; In brief, this lung feature is comparable with typical feature of tuberculous tubercle in both lungs in man, although fungi-elements are lacking in the latter tubercle.

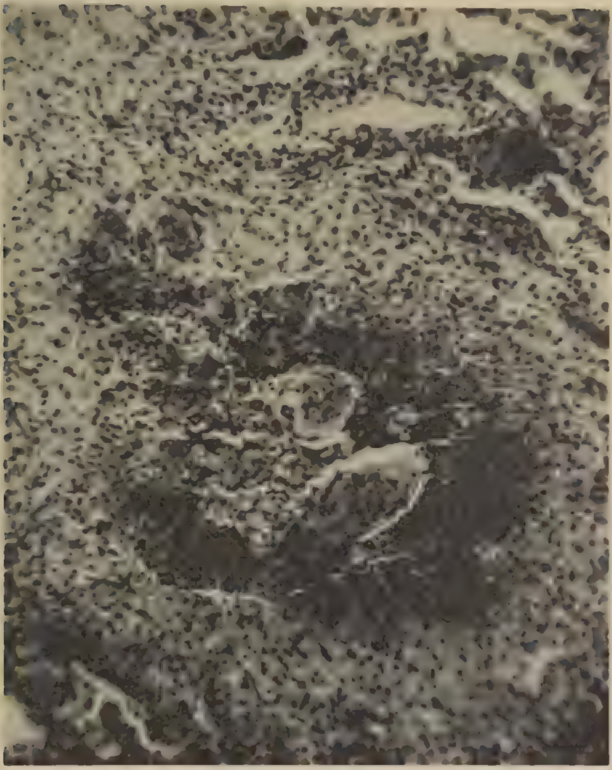


Fig. 8. Showing giant cell of Langhans type contained in the tuberculoid granuloma. The giant cell phagocytizes damaged fungielements (as stained by Mc Manus method), lymphocytes and damaged polymorphs with nuclear débris (specimens were taken from left lung of rabbit destroyed 3 weeks after the intravenous reinjection of tr. ast. suspension. The fungus granuloma had the central necrotizing area (damaged fungi-elements plus damaged polymorphs accumulated with nuclear débris). The fungus granuloma was produced in lungs of rabbit due to the intravenous reinfection with the tr.

IV. Evaluation of crude polysaccharide (from the tr, mentagr., var. ast. mechanically disintegrated) as antigen (used for serology).

The importance of serology in the field of mycosis including mycid is beyond question. Unfortunately, much remains to be answered in this field, because of the lack of suitable antigen. We have prepared as this antigen crude fractions (crude polysaccharide in particular) from fungi mechanically disintegrated.

The disintegration is performed using the special disintegration apparatus in our institute. Our experiences teach us an excellent antigenic activity of the said fungus fraction in experimental trichophytia (inclusive of fungus granuloma and other fungus "id") as well as in patients with dermatomycosis (including mycid), while relatively refined polysaccharide from the tr. (not mechanically disintegrated) proved to be not so sensitive in reacting to antibodies as was the crude polysaccharide above given.

Also, attention is called to the fact that the experimental production of the fungus granuloma (on the ears of rabbits) approximately coincided with the serological flare up in the rabbits — The flare up followed second dorsal inoculation with the tr. ast. in the rabbits, which had been sensitized by dorsal inoculation with the tr., but antibodies against the said crude polysaccharide had decreased or disappeared their production a certain length of time after the "healing" of the first inoculation on the dorsum.

V. The use of crude polysaccharide (from fungi mechanically disintegrated) as trichophytin.

Using crude polysaccharide (from tr. mentagr., var. ast.) as trichophytin, the trichophyt in skin reaction was demonstrated in experimental trichophytia of rabbits, while relatively refined polysaccharide (from the tr. not mechanically disintegrated) did not produce such intensive trichophytin reaction as the crude polysaccharide above given. Particular attention is called to the flare up of trichophytin (the said polysaccharide) reaction in rabbits. This flare up was observed when the fungus granuloma was produced on the ears upon a dorsal second inoculation with the tr. ast. These reinoculated animals also showed serological flare up (in IV given)— This humoral reaction was demonstrated, using as antigen the same crude polysaccharide trichophytin. In short, a combination of trichophytin (crude polysaccharide from the tr. mechanically disintegrated) reaction with serological reaction (using the same crude polysaccharide as antigen) will provide an useful proof for the pathogenesis and diagnosis of fungus granuloma experimentally produced.

Summary

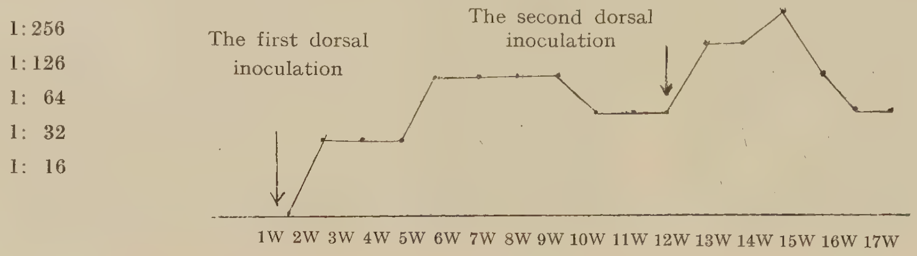
Here is presented a case of the experimental production of fungus granuloma on the ears of rabbits due to a second dorsal inoculation with tr. mentagr., var. ast. This tuberculoid granuloma is preceded or accompanied by fungus phlebitis and occasionally associated with arteritis or perifolliculitis granulomatosa on the ears. These reactions (granuloma, phlebitis, arteritis and perifolliculitis) are regarded as almost certainly falling into a cathegory of trichophytid as demonstrated by histology, serology, trichophytin and blood-cultivation. Also, these reactions are comparable to the cases of human fungus "id" reactions and tuberculous reinfection lesions in man. Then, similar fungus granulomas were artificially

TABLE I.

Trichophytin** skin reaction and humoral response*** in the rabbit, in which the fungus granuloma* was produced on the ears upon a second dorsal inoculation with tr. mentagrophytes, var. asteroides.

Item/Time		1W	2W	3W	4W	5W	6W	7W	8W	9W	10W	11W	12W	13W	14W	15W	16W	17W
Skin manifestation	Redness****	+	+	+	+	+	+	±	—	—	—	—	+	⧺	+	—	—	—
	Induration****	+	+	⧺	⧺	+	+	±	—	—	—	—	+	⧻	+	—	—	—
	Cruste****	±	+	⧺	⧺	+	+	±	—	—	—	—	+	⧻	+	±	—	—
	Fungus-granuloma*****	—	—	—	—	—	—	—	—	—	—	—	—	+	+	+	+	+
	Corymbiform eruption******	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
	Generalized papular lesion	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Skin reaction with	Crude polysaccharide	—	—	+	+	⧺	⧺	+	+	+	+	+	⧺	—	⧺	—	—	+
	Crude lipid	—	—	—	—	+	+	—	—	—	—	—	+	—	+	—	—	—
	Crude protein (on the ear)	—	—	—	—	—	—	—	—	—	—	—	+	—	—	—	—	—

Titer of Antibody



→ Weeks after the dorsal inoculation with tr. mentagrophytes disintegrated

— indicates complement fixation tracing.

* centering around fungus abscess.

** prepared from tr. mentagr., var. ast. mechanically disintegrated.

*** used as antigen crude polysaccharide (from tr. mentagr., var. ast. mechanically disintegrated.)

**** on the back of the rabbit caused by the primary dorsal infection with tr. mentagr., var. ast.

***** on the ear of the rabbit caused by the dorsal reinfection with tr. mentagr., var. ast.

****** on the dorsum of the rabbit caused by the dorsal reinfection with the tr.

produced on the ears and in both lungs etc. by disseminate reinfection with the tr. ast. via cardiac route in rabbits, which had immunized by a dorsal infection with the tr. Summed up, this work is concerned with experimental fungus granuloma as seen from comparative exanthematology of mycid with other "id", tuberculid in particular. We consider the fungus allergic granulomatous reactions as partial manifestations of general immune reaction (including trichophytemia), regardless whether fungi in the lesions activate in loco or result from fungus foci via blood circulation. Also, emphasis is placed upon an evaluation of crude polysaccharide (from the tr. mechanically disintegrated) as antigen or trichophytin (used for serology or trichophytin reaction).

References

Brocq-Rousseu, Urbain, A. & Barotte, J. 1927. Anticorps dans les teignes experimentales: Zbl. Haut- u. Geschlechtskr. **21**: 853.

Brunsgaard, 1922. Über hämatogene Trichophytie: Arch. Dermat. & Syph. **138**: 381.

Bukovsky', J. 1900. Ein Beitrag zur Kentnis der experimentellen und klinischen Eigenschaften des Achorion Schönlein: Arch. Dermat. & Syph. **51**: 365.

Falchi, G. 1927. Trichophytid mit Kultur des Tr. plicatile in strömendem Blut: Derm. Z. **18**, 891.

Fried, M. & Segal, B. 1928. Beiträge zur experimentellen, auf hämatogenem Wege hervorgerufenen Dermatomykose, mit Berücksichtigung der Zirkulationsdauer der Parasiten im Blutkreislauf: Derm. Wschr. **87**, 1426.

Henrici, A. T. 1939. Experimental trichophytid in guinea pigs: Proc. Soc. exp. Biol. Med. **41**, 349.

Jadassohn, W. 1927. Beitrag zur Genese der Allergie bei Impfmykosen: Arch. Dermat. & Syph. 153: 476.

Jessner, M. 1921. Zur Pathogenese der Thrichophytide: Arch. Dermat. & Syph. **136**: 416.

Kogoj, F. 1926. Experimentelle Beiträge zur Lehre von den Dermatomykosen mit besonderer Berücksichtigung der Lokalisations-bestimmung hämatogener Infektionen: Arch. Dermat. u. Syph. **150**: 333.

Kogoj, F. 1928. Hämatogene Dermatomykosen: nach subkutaner und intramuskulärer Infektion: Arch. Dermat. u. Syph. **154**, 463.

Saeves, I. 1916. Experimentelle Beiträge zu Dermatomykosen: Arch. Dermat. u. Syph., **121**: 161.

Smolka, 1928. Experimentelle Beiträge zur Frage der hämatogenen Dermatomykosen: Arch. Dermat. u. Syph. **154**, 519.

Sulzberger, M. B. 1928. The Pathogenesis of Trichophytids: Arch. Dermat. a. Syph., **18**, 891.

CRITICAL SURVEY OF MEDICAL MYCOLOGY IN CZECHOSLOVAKIA FOR THE YEARS 1946 TO 1956

by

LADISLAV CHMEL, M.D.

Physician in charge of the Dermato-venerologic Clinic, Bratislava, Czechoslovakia

(5.VI.1959)

At the very start of a survey covering the incidence of dermatophytoses on the territory of Czechoslovakia it is necessary to draw the reader's attention to the fact that two distinct regions will be considered here: Bohemia and Slovakia. These two regions differ not only geographically but to a certain extent economically and socially as well.

Hrad. Králové

The geographical difference originates from the character of the Bohemian lands which consist of hills broken by plains and from the special character of Slovakia typical by its high mountain ranges in the North and the West and by low plains extending in the South of the country. Economical and social differences, once upon a time very marked between Bohemia and Slovakia, are gradually disappearing but their consequences are to some extent being felt to the present day. If we add to this the economic damage caused by the passage of the war front through the East territory

(Slovakia) of the State with the consequent deterioration of the sanitary situation in these regions as a whole, we find an explanation not only for the number of ailments of the dermatomycosis type and for the qualitative differences of the causes of such diseases, but also for the differences in the epidemiologic situation in these two parts of Czechoslovakia.

Deep mycoses occur in this country only in exceptional cases. That is why in this survey we limit our data to dermatomycoses caused by filamentous fungi.

After World War II Czechoslovak dermatologists in accordance with the general policy of prevention followed in the organization of public health set to wage war against dermatomycoses. Their work performed directly in the field, and where necessary, in close cooperation with the veterinary services, has acquainted them with the laws of the epidemic process and helped them to fathom the question of the epidemiology of dermatomycoses in this country. The question of trichophytoses, microsporosis, favus and of epidermophytoses has gradually assumed important proportions and its study has become the object of the labour of many a scientific worker.

The first studies dealing with this problem were published by CHMEL after the war. His publication (5) issued in 1946 gives an outline of the composition and of the dynamics of the dermatophyte flora in the Western part of Slovakia in the various years ranging from 1942 to 1946. This analytic work was followed by another of a purely practical tendency (6) published by the same author in 1947. Its aim was not only to help the practician in the diagnosis of dermatomycoses, but give him the necessary advice and an outline of the campaign for the realisation of preventive steps to be taken in case of an increasing incidence of dermatomycoses. In 1948 CHMEL published another study (7) dealing with the importance of preventing dermatomycoses in the armed forces, stressing the importance of prevention of epidermophytoses which are most common among the armed forces. The same point — seriousness of the disease and the importance of preventing the same — is stressed in his next work published in 1949 (8) where he deals also with the question of epidermophytoses among sportsmen. The first signs of collaboration between the physician, the veterinary doctor and the sanitary officer are outlined in the works of CHMEL and his collaborators (9) published in 1950 and those of NIŽNÁNSKY and his collaborators (70) published in the same year.

These first studies connected with human medicine published after the war and in which a general remark of the increase of dermatomycoses was made, are followed by the works of NIŽNÁNSKY (71, 72) published in 1952 and 1953. In these studies he deals with the incidence of trichophytoses in cattle; at the same time he draws attention to the necessity of mustering the whole veterinarian corps of Czechoslovakia into the struggle against these diseases.

In the year 1953 CHMEL in his work (10) entitled "Some actual problems in the struggle against dermatomycoses in our country" suggests the measures to be taken as a basis for the organisation of a systematic campaign against dermatomycoses. The question of organized struggle against dermatomycoses is further developed in 1954 by JANDA (62) dealing with the preventive measures against these diseases and again by CHMEL & HEGYI in 1955 in their "Notes on the organized struggle against dermatomycoses in the country" (14) where the authors give concrete suggestions and state the fundamental principles along which the fight against dermatomycoses is to be carried on under given, relatively limited possibilities.

At this time we witness the publication of the first studies treating with the composition and dynamics of the dermatophyte floras in the various regions of the country above all the agents provoking trichophytoses, microsporosis, favus and partly of epidermophytoses.

As regards the incidence of dermatomycoses it can be said that far more mycotic diseases provoked by dermatophytes occur in Slovakia than in Bohemia. There is also a difference in the composition itself of the dermatophyte flora.

Dermatophytes are cultivated in several laboratories of the country but especially in the medical faculty laboratories established in the proximity of dermatologic clinics.

The results of mycologic cultures from larger laboratories published in the period 1946—1956 give an idea of at least the main lines of the composition of the dermatophyte floras on the territory of Czechoslovakia in the period under revue.

The most abundant data relating to the period 1946—1956 were gathered by CHMEL from the mycological laboratory of the Dermatological clinic in Bratislava. A research lasting several years and carried on together with 19 dermatologists and partly also with a number of veterinarians from all the regions of Slovakia brought in some very extensive material. This material though it does not give an exact idea of the number of cases of trichophytosis, microsporosis and favus, which were the primary object of the research (the infectious material was taken only from certain infected cases or only from part of the collective in which the epidemic was) it nevertheless enables us to have a more definite idea about the dermatophyte floras of the regions of Slovakia. This study follows on the publication (5) giving the results of mycologic cultures at the dermatologic clinic in Bratislava in the years 1942—1946.

TABLE I.

Dermatophytes isolated in the regions of Slovakia during the period 1946—1956

Trichophyton gypseum	955	*Microsporum audouini*	104
,, *niveum*	3	,, *ferrugineum*	2
,, *persicolor*	1	,, *gypseum*	9
,, *faviforme*	439	*Achorion schönleini*	26
,, *rosaceum*	14	,, *quinckeanum*	1
,, *violaceum*	74	*Epidermophyton Kaufmann-Wolf*	105
,, *sulfureum*	10	*Epidermophyton rubrum*	55
,, *crateriforme*	7	,, *floccosum*	23
,, *cerebriforme*	3	*Candida albicans*	291
,, *acuminatum*	1	,, *guillermondi*	14
		,, *brumpti*	2
		,, *krusei*	5
Total			2.184

A partial survey of the composition of dermatophytes from patients treated at the Dermatologic clinic in Košice in the years 1946—1954 is given by KACHNIČ (65):

Trichophyton faviforme	26
Tr. gypseum aster.	24
Tr. gypseum granulosum	6
Tr. violaceum	10
Tr. crateriforme	1
Achorion Schönleini	1
Epidermophyton inguinale	1
Microsporum audouini	1
Total	70

GODOVIČ (40) gives a similar table from the region round Michalovce, where in the period from December 1952 to June 1955 200 culture-proved cases of dermatomycosis occurred, to wit:

Trichophyton gypseum asteroides	73 %
Tr. faviforme	20 %
Tr. violaceum	3 %
Achorion gypseum	2.5 %
Microsporum audouini	1.5 %

From the regions of Bohemia we have no such outline of the composition of dermatophyte flora as we have from Slovakia. Some idea may be gathered from the work of GROH (42) who gives a summary of the results of cultures in the region of Hradec Králové during the years 1946—1950:

Trichophyton mentagrophytes	75
Tr. epilans	74
Tr. sabouraudi	1
Tr. rosaceum	19
Tr. schönleini	5
Tr. violaceum	11
Microsporum audouini	8
Epidermophyton floccosum	1
Candida albicans	31
Total	225

Sedláček (83) from the dermatologic clinic of Brno gives a table of the results of mycologic examinations for the regions of Moravia in the years 1946—1951:

Trichophyton gypseum asteroides	98
Tr. rosaceum	13
Tr. violaceum	5
Tr. cerebriforme	2
Tr. inconnus	6
Achorion schönleini	3
Ach. quinckeanum	4
Microsporum audouini	1
Epidermophyton Kaufmann-Wolf	12
E. inguinale	2
Total	146

From the data given here we may conclude that cases of dermatomycoses occur substantially in greater numbers in the Slovak regions than in those of Bohemia and that in the composition of the dermatophyte floras there is also a qualitative difference, namely that in the Slovak regions among the dermatophytes of the group *Trichophyton*, *Microsporum* and *Achorion*, the zoophilic dermatophytes are much higher in number than the anthrophilic dermatophytes (Chmel 83,5 : 16,5 %, Kachnič 80 % : 20 %, Godovič 95,5 % : 4,5 %) thus differing from the regions of Bohemia where according to Groh their ratio is equal or is in favour of anthropophilic dermatophytes.

Chmel (20) gives records of the relative representation of the cases of trichophytic, microsporic and favic diseases in the group of mycoses provoked by dermatophytes (epidermophytons excepted). From 1894 cases of dermatophytes recorded by him in the period of 1952—1955 in 1806 patients, i.e. in case of 95,3 % there was question of trichophytic infection, in the case of 68 patients i.e. 3,6 % there was Microsporosis and 20 patients, i.e. 1,1 % had favus.

At the same time some dermatologists began to devote a closer attention to the problem of epidemiology and to analyse more thoroughly various epidemics, often directly in the field. They have thus gained data for well aimed antiepidemic measures. The following studies belong to this group: "Contribution to the problematics of dermatomycoses" by Šteklác, Drgonec & Ružička (88) published in 1953 and in which the authors analyse the cases of dermatomycoses treated at the dermatological section of the military hospital in the years 1951—1952. The work of Dvořák from the same year (23) gives the results of the medical examinations for dermatomycoses done in connection with military service. The work of Langer (67) from 1953 deals with a fairly considerable epidemic in a children's group, provoked by an imported strain of *Microsporum ferrugineum*. The work of Pálešová (77), 1953, which follows up the question of the incidence of favus in the region of Bratislava from the point of view of the epidemiology of these diseases and suggests a basis for eventual antiepidemic measures. CHMEL & BABÁK in 1954 (11) in a clinical and mycological study describe a case of superficial trichophytosis provoked by the strain of *Tr. sulfureum*, a dermatophyte which was first obtained by culture in Czechoslovakia. Further, an epidemiologic study by CHMEL & PÁLEŠOVÁ (12) from the year 1954 concerning 38 cases of favus recorded at the dermatological clinic of Bratislava from the year 1940 to 1953 of which only 7 cases did not come from Slovakia (D.P. from Hungary). The analysis shows that while in the regions of Bohemia and Moravia favus is a rarity, it is much more frequent in Slovakia.

The work of CHMEL (16) (1956) is a thorough epidemic study dealing with the question of the importation of *Tr. rosaceum* to the territory of Czechoslovakia and the incidence of extensive epidemics which it had provoked and which were caused by the retreating Nazi soldiers at the end of the Second World War. Part of the study is devoted to the clinical study of these diseases caused by this dermatophyte in so far as they were treated at the Bratislava skin clinic.

The works of SEDLÁČEK (86), 1955 and that of HORÁČEK & LANGER (56) (1956) are epidemiologic studies of mycotic infections in children's groups. The first deals with the epidemic caused by *Microsporum ferrugineum*, the second with the mass incidence of trichophytosis caused by *Tr. sulfureum*.

From the preceding works it can already be seen that the incidence of dermatomycoses gravitates towards agriculture. In 1956 a more specialized study of the relation between professional diseases and agriculture was carried out by ERBÁK (28), VIŠČOR (91), FRÁGNER & KRAUSKOPF (32) and in the next year by GODOVIČ (40), HÜBSCHMANN, KRAUSKOPF & FRÁGNER (58), CHMEL & PÁLEŠOVÁ (17) and CHMEL (20, 21).

ERBÁK in his work (28), illustrated by the results of culture studies, analyses the incidence of epidemics and epizootics of the region falling under the jurisdiction of the skin section of the Hospital at Rimavská Sobota. He devotes a special attention to the State farms, Farmers' Cooperatives and private farmers, i.e. large farms and smaller ones.

VIŠČOR (91) gives an analytic outline of the results of a research done in the chief centers of animal production at the State farms and at farmers' Cooperatives in the region of Turiec in the year 1952—1953. This analysis gives practical information for the fight against dermatomycoses.

Similarly FRÁGNER & KRAUSKOPF (32) as well as HÜBSCHMANN, KRAUSKOPF & FRÁGNER (58) give a report of the incidence of epidemics and of epizootics in the agricultural bases of the Prague region where in 85 % of the cases trichophytic infection provoked by *Tr. faviforme* or *Tr. gypseum* was found among the employees.

GODOVIČ (40) discusses the problematics of a higher incidence of trichophytoses in East Slovakia from the point of view of the information obtained from the dermatological section in Michalovce. In this work resulting from field studies he analyses the different conditions of the spread of trichophytic infections caused by zoophilic strains in the agricultural centers of the socialist sector on the one hand and in the private sector on the other. He shows also the different possibilities of fighting against epidemics in both of these sectors.

CHMEL & PÁLEŠOVÁ in their work (17) give their experience of field work. They point to the seriousness of the incidence of dermatomycoses in agriculture and show that it would be possible to prevent the damage resulting from so many labour hands being disabled from work as well as from often widespread epizootics, could be prevented by the right kind of coordination among dermatologists, veterinarians and sanitary officers.

The most extensive and at the same time the most complex studies of the epidemiology of dermatomycoses in agriculture, describing the conditions in the regions of Slovakia, are the publications by CHMEL (20, 21). Together with all the dermatologists

TABLE II.

Distribution of trichophytoses, microsporosis and favus according to the avocation of the patients ¹)

(for the period 1.XI.1952—31.8.1955)

	Trichophyt.		Microspor.		Favus		Total of dermatomycoses	
	Number	%	Number	%	Number	%	Number	%
Agricultural workers	813	79,8	—	—	4	50,0	817	76,4
Industrial workers	65	6,4	7	16,6	1	12,5	73	6,8
Clerks	82	8,0	3	7,1	2	25,0	87	8,1
School children ²)	28	2,7	31	74,0	1	12,5	60	5,6
Housewives and others	32	3,1	1	2,3	—	—	33	3,1
Total	1020	100,0	42	100,0	8	100,0	1070	100,0

¹) Children under fourteen are classed according to their parents' avocation.

²) Not taking into account the parents' avocation.

of Slovakia he carried out, during the period from 1.VI.1952 to 31.VII.1955 a systematic study of the epidemiological situation centered on the incidence of dermatomycoses in agriculture. He was led to it by the finding which showed that not only 76,6 % of all the dermatomycoses (trichophytoses, microsporosis and favus) are centered in agriculture, but also 79,8 % of all recorded trichophytosis are found there as is clearly seen from the forgoing table which gives a survey of the distribution of trichophytosis, microsporosis and favus according to the avocation of the patients.

In connection with this study the author follows up the dynamics of the incidence and spread of trichophytosis provoked by zoophilic dermatophytes.

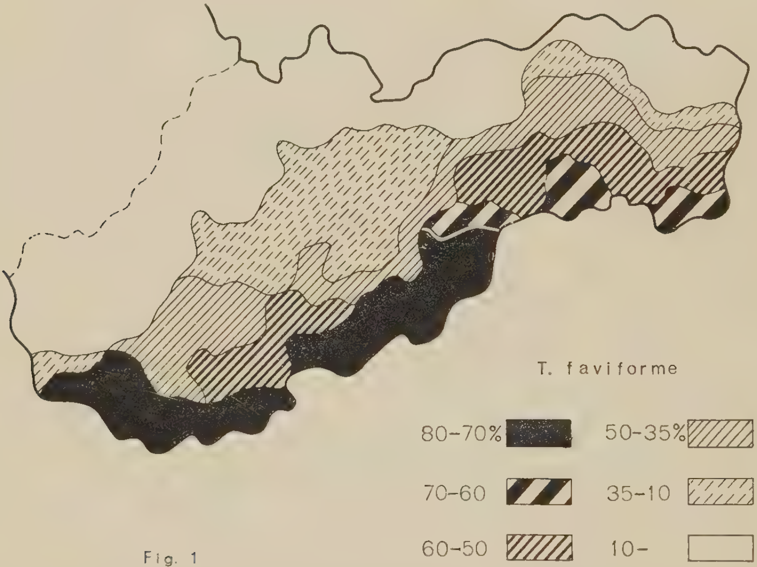


Fig. 1

Cultures in dermatomycoses from infectious material taken from persons working in agriculture has shown that from a total number of 642 positive cultures in 621 cases (96,7 %) there was question of zoophilic strains and in 21 cases (3,3 %) the cause of dermatomycoses was the anthropophilic dermatophyte. Out of 621 instances 618 cases were caused by *Trichophyton strains* (*Tr. gypseum* in 396 cases, *Tr. faviforme* in 218 cases and *Tr. rosaceum* in 4 cases), in 2 cases there was question of *Microsporum gypseum* and in one case of *Achorion quinckeanum.*

Figures 1 and 2 show the geographical distribution and the percentage of *T. faviforme* and *T. gypseum* from the total number of these two strains isolated in the course of three years here considered. One does not fail to notice at first glance the striking difference between the existence of *T. faviforme* which is mostly found in the plains of Slovakia only (80 %) and the existence of *T. gypseum* which is found above all (90—100%) in the mountainous Karpatian regions.

It follows from this that the geographical differences between these mountainous regions on the one hand and the plain on the other lead to differences in the character of the life-stock economics, in the working conditions as well as in the character of the epidemic processes of dermatophytoses.

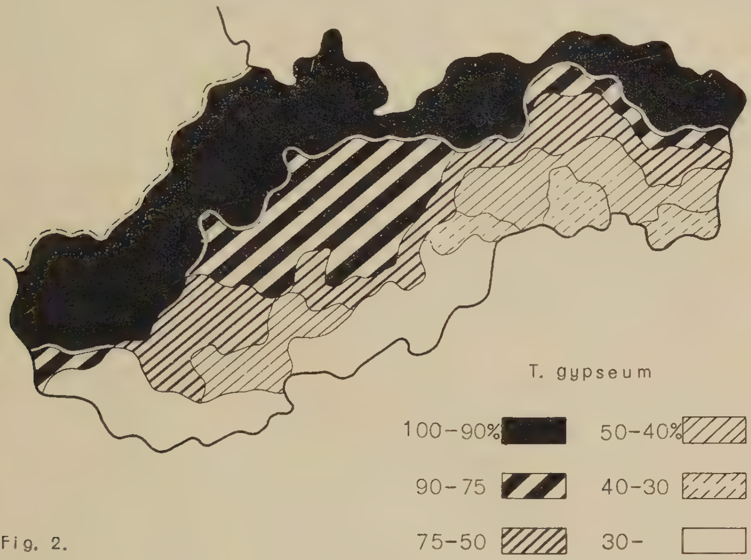


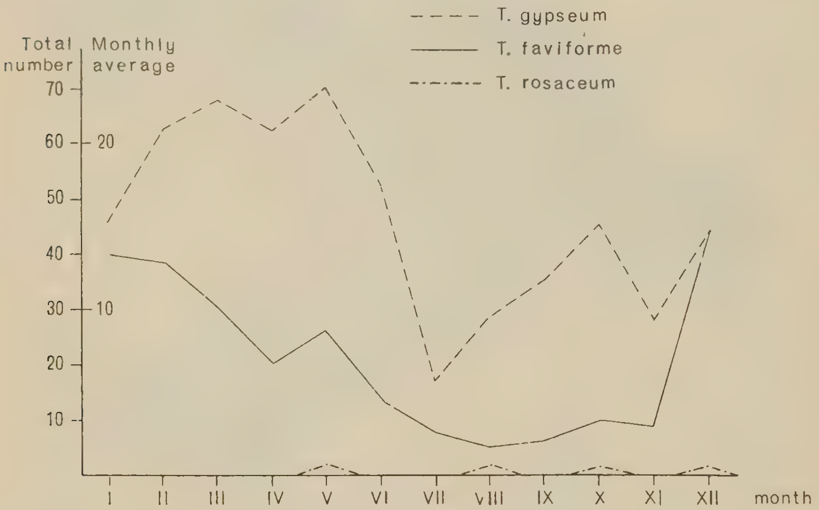
Fig. 2.

These differences were evident already from the indications given by the patients themselves. In 269 cases of dermatophytoses caused by *T. faviforme* life-stock diseases were given as source of infection. In 27,9 % the source of infection was not given and could not be traced. On the other hand in 427 cases of dermatophytoses caused by *T. gypseum* only in 7,6 % of the cases was infected cattle given as source of contamination. These figures show clearly that the chief host of *T. faviforme* are cattle whereas the hosts of *T. gypseum* are rodents with which humans do not come into direct contact.

It is interesting, besides, to note which months of the year abound most in trichophytic infections, what the extent of these infections is (Graph 1) and what is the relation between the infections caused by *T. faviforme* and *T. gypseum* in large cattle-rearing farms as well as in smaller ones. In the former *T. faviforme* is present in 62,2 % and *T. gypseum* in 32,2 % of the cases of disease; in the latter *T. faviforme* represents 14,6 % and *T. gypseum* 85,4 % of the cases of disease.

Cattle rearing on a large scale is carried on above all in the plains of Slovakia. Accommodating cattle in stables, without pasture, without yards where the animals may take exercise contributes to spread diseases caused by *T. faviforme*, not only among the cattle but also among labour hands employed in stock production (Graph 2).

On the other hand, heat, humidity (dampness and the half-obscurity of the stables make favourable conditions for the growth of dermatophytes elsewhere than on macroorganisms (such as dung, hay, straw etc.) which fact is not without importance in the transference of infection to man.

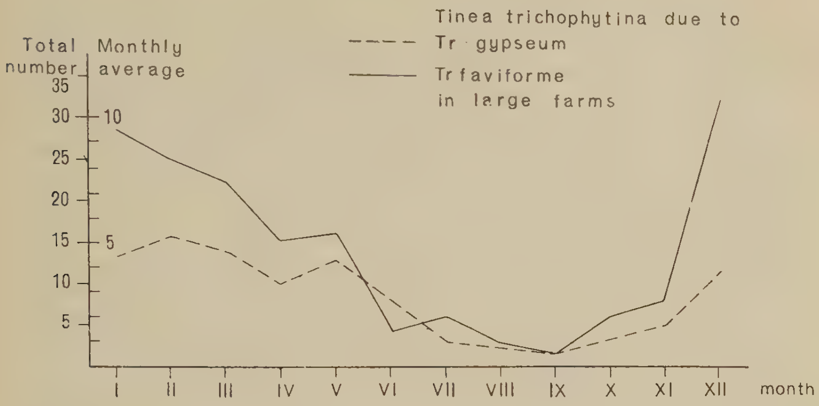


Graph 1.

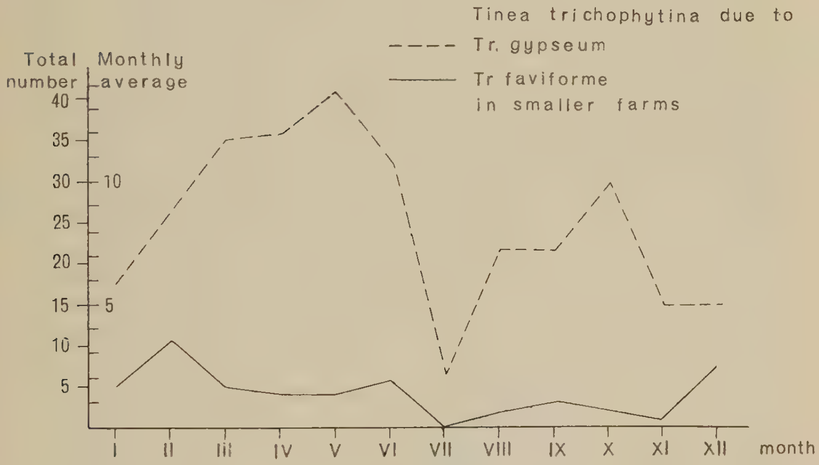
In the mountainous regions few trichophytic diseases are met with. This may be explained by the smaller number of animals — in many cases it is a question of only one or two head of cattle per farm — as well as by the fact that in the summer months the cattle frequent the pastures and enjoy the sun and the open air (graph 3).

With epidemiologic and epizootologic circumstances taken as base we can explain also the increase as well as the decrease of trichophytic diseases caused by *T. faviforme* in the various seasons of the year.

The lowest number of cases is connected with the long stay of cattle in the sun and in the open air. The increasing frequency of diseases at the end of autumn (with peak numbers in December) results from the prolonged stay of the cattle in stables as well as from the closer contact of the farmer with the cattle in winter. Under the influence of treatment and under the effect of improved



Graph 2.



Graph 3.

climatic conditions we note, with the coming of spring, a decrease in the number of cases of disease. The temporary increase in the number of these diseases in May may be explained by the transmission of infection by means of infected hair which the animal shed at this time of the year. (Graph 1).

The reason of the incidence of *T. gypseum* in the mountainous regions rest in the fact that in the houses where annexe buildings are connected with the house, these buildings are inhabited not only by mice but also by other rodents who come for shelter from the neighbouring fields and woods. In general, in mountainous regions wooden structures are easily accessible to mice and the spaces between the roof planks are suitable places where rodents naturally make their abode.

In the plains where the houses are stronger and are built of brick and stand apart from human habitations the mice can less easily approach human dwellings. Besides, large, almost limitless areas of fields do not offer the mice favourable life conditions.

Mice are timid by nature and the dermatophytes are transferred from them mainly indirectly. Therefore here the survival of dermatophytes outside the host, on the straw, on fodder and on various household utensils plays a fairly important role.

What has just been said explains also the increase and decrease in the number of cases of disease in connection with the various seasons of the year. The increase in August and September is explained as a result of a closer contact of men with mice during the harvest. The second increase in winter months is linked with the life of mice in human dwellings or close to them. Lastly the maximum number of cases appearing in April and May is explained by the utilisation of the lower layers of straw, hay and other stocks which had been mostly frequented by mice in winter and where the greatest amount of contaminated stuff such as hair, dung and other infected material is left by them. The sudden decrease in the number of cases in the summer months follows from the migration of mice into the fields and from the exhaustion of fodder stocks.

From the preceding analysis follows the specificity of procedures evoked by the individual kinds of zoophilic trichophytons. It clarifies the relation existing between the source of these infections and the diseases resulting from them. This analysis will also be the starting point of specific prophylactic steps in which account is to be taken not only of the biology of the type of dermatophyte and his main host but also of natural, economic and social conditions under which the farmer is obliged to live.

HÜBSCHMANN & FRÁGNER (61) describe and analyse the Candidosis interdigitalis of goose-feather pickers as a new professional dermatosis provoked by *Candida albicans.*

Epidemics of dermatomycoses in connection with avocations have been followed up in industrial factories, too.

HEGYI, KUŽELOVÁ & LENDVAI (44) describe (1956) the incidence of an epidemic with symptoms of epidermophytosis corporis in a brush factory in persons working with bristles. The causing agent was *Tr. interdigitale Kaufmann-Wolfi.*

Similar in character is the study of FRÁGNER & KRAUSKOPF (35) (1955) dealing with the incidence of epidemic caused by *Tr. gypseum* var. *Quinckeanum* in the chantier of a factory where the workers worked with straw. 11 out of 17 workers were affected by superficial dermatomycoses.

The works of HÜBSCHMANN et al. (57, 59, 60) also have an epidemiological significance. These studies are an attempt at determining an average picture of the mycotic flora of normal epidermis.

It is not always easy to distinguish studies of an epidemiologic character from works having a prophylactic character. A considerable portion of epidemiological studies usually contain in their final stages a sketch of preventive measures. The outline of epidemiology and of the epidemic situation in Czechoslovakia as well as studies giving concrete suggestions as to the methods of carrying out prophylactic actions were certainly a welcome foundation for the drawing up of instructions for the struggle against dermatomycoses; this was the basis for the realisation of the Act of the Ministry of Health of 1955 making it compulsory to fight against trichophytosis, microsporosis and favus and of reporting their incidence to public health authorities.

Of such an organizing character are the works of Chmel who apart from the above mentioned work (6) deals in a special study (10) published in 1953 with certain actual problems of the struggle against dermatomycoses. In collaboration with Hegyi (14) in 1955, starting from the information gained in the field work in connection with a survey in agriculture (17), he deals with the question of the fight against dermatomycoses especially in the centre itself of these disease, i.e. in the country. In further studies (15) (1956), (19) (1956) Chmel developed theoretically the principles of epidemiology and prevention of dermatomycoses much in the same way as Janda (62) (1954), (63) (1956) and Hegyi (45) in 1956. The work of Dvořák & Výmola (27) published in 1956 deals with the question of prevention of candidoses in antibiotical treatment. On the veterinary field this question is dealt with by Nižnánsky (70, 71, 72) in the years 1950—1953.

Quite a number of authors deal with the problem of diagnosis, laboratory as well as clinical.

Sedláček in his work (81) published in 1950 deals generally with the question of prevention, including the diagnosis and therapy of dermatomycoses most often met with in praxis. Differential diagnosis and therapy of oidio- and dermatomycoses is the theme of Obrtel's work (74) published in 1949. To this group of studies dealing with clinical diagnosis belongs also his work (75) from 1950 dealing with "ids" in dermatology, as well as Sedláček's casuistic (82) from 1951 about the case of balanoposthitis caused by *Candida*, Vacátko's casuistic (90) of 1956 dealing with a case of Favus squamosus and Dvořák's (26) of 1956 about the incidence of erythrasma in intradigital spaces on foot.

Sedláček in his work (80) of 1950 devotes himself to the study of trichophytin, summarising his experiences with trichophytin of his own production. Langer (66) in 1950 deals with the question of standardization of trichophytin and Hegyi (43) in 1954 makes a study of the hypersensitivity to trichophytin from the point of view of the diagnosis and the therapy of epidermophytides and evaluates the significance of an early and late reaction to this product with a view to practice. Liška (68), (1956) attempts a quantitative and qualitative determination of hypersensitivity to trichophytin with the help of iontophoresis.

Hegyi, Chmel & Martin (46) in 1957 deal with the possibilities of ameliorating the conditions of diagnosis in mass examinations in the field. They suggest a simplified apparatus for luminiscence as well as a simplified black-out system suitable for every situation.

The work of Balun (1), 1956, dealing with microcultivation of dermatophytes and the work of Groh (41), 1951, concerning the use of penicillin and streptomycin in the isolation of dermatophytes are suggestions for better laboratory examinations. Hejtmánek's study (48), 1956, is a contribution to the study of the morphology of dermatophytes in an electron microscope. Obručník's publication (76), 1953, is a histological study of the changes in the structure of the epidermis of interdigital spaces on the human foot and of their relation with the pathogenesis of skin diseases of these regions.

Quite a number of publications shows an interest in the study of antimicotically effective materials.

Horáček (55), 1948, and Polster (78), 1951, make a study of the fungicidal effect of the derivates of phenol. Polster, besides, makes a study of the mechanical effect of 2-chlor-4-nitrophenol in certain enzymatic reactions of *Epidermophyton Kaufmann-Wolf* (79), 1954, and examines the complete inhibition in the case of intracelular nitritereductasis and of TTC-dehydrogenasis.

Kabelík (64), 1954, gives an extensive study of anorganic fungicidal materials from which the most suitable in praxis proved mercury and copper salts, including organic salts. The most effective proved to be nitroderivatives — chlornitrophenols.

Nosek & Dvořák (73), 1954, give the results of the antimycotic effect of etylenoxyde and of its handiness in the disinfection of military equipment. Sedláček (85), 1954, using the method of filter paper follows up the antimycotic effect of a whole range of organic and anorganic materials and of a group of 71 various vegetable oils. Frágner et al. (31, 33, 34) study the antimycotic effect of isofurancarbonic acid, of chinosol and certain derivates of chinosol. Uličná (89), 1956, in a short study gives the results of the effect of chlornitrophenol on *Candida albicans* and *Achorion Schönleini*.

Finally Chmel (18), 1956, examines by "agar-cup plate" method the antimycotic effect of seven disinfecting preparations produced in Czechoslovakia ¹) with a view of their practical ap-

---

¹) Ajatin-Synfarma (dimethyllaurylbenzylammonium chloratum),
Famosept (Ryfen)-Synfarma, SPOFA (Phenylhydrargyrum boricum),
Fenol-ČsL 2 (Fenolum),
Chloramin-Spolana (Natrium p-toluensulfochloramidicum),
Chlorseptol-Spolana (Natrium p-toluensulfonchloramidicum with addition of azo-orange),
Kreolin-Slovakofarma (mixture of kresol-57 % and soap-16-17 %),
Liquor kresoli saponatus-Slovakofarma (50 % kresoli and 50 % saponis kalini).

plication in the struggle against dermatomycoses. In this study he examines the order of merit and the degree of effectiveness of the individual prophylactic materials as well as the order of merit and the degree of the general and individual sensitiveness of pathogenic fungi. Of 18 different strains examined here the most sensitive to prophylactic material proved to be *Microsporum Audouini*, *Tr. violaceum*, *Tr. faviforme*, *Ep. floccosum* and *Achorion quinckeanum*. Of average sensitiveness are *Tr. crateriforme*, *E. Kaufmann-Wolf*. The least sensitive to prophylactic materials proved to be *Tr. gypseum aster.*, *Microsporum gypseum* and above all *Penicillium* and *Candida albicans*. In the study a special attention is given to the dynamics of antimycotic effects of the material observed with regard to their degree of concentration.

HORÁČEK (54), 1946, deals with the problem of the treatment of sycoses and gives the results of his experiences. SEDLÁČEK (84), 1952, gives the results of treatment of trichophytoses with CO_2 snow (carbonic snow). DVOŘÁK & DOČEKAL (24), 1953, describe their experiences with the treatment of dermatomycoses with formic acid. BENCA (2), 1954, speaks of his experience with the treatment of deep trichophytoses with podphylin. FABIAN (29), 1956, gives an outline of the results of treatment of microsporosis which were met with in the region of Ostrava and BLATICKÁ (3), 1956, gives a note on the methods of treatment of dermatomycoses in patients at the dermatologic clinic of Košice. Finally NEUMANN (69), 1956, as well as ŠT'ÁVA & JIRÁSEK (87), 1956, follow up the experience with the use of hyaluronidase, with the help of which the nail is removed before the antimycotic treatment of the nail cavity.

Finally this last part may include all the other studies which did not directly find their place in the previous classification.

They are in the first place studies dealing with the biology of dermatophytes. CHMEL & PÁLEŠOVÁ (13), 1954, follow up certain manifestations of variation in the organism of favus. Further, a whole range of studies of HEJTMÁNEK M. & HEJTMÁNKOVA N. from the period 1955—56 could find their place here. They deal with the variability and the polymorphy of dermatophytes (47, 49, 50) as well as the study of the origin of coremia in cultures of *Tr. Kaufmann-Wolf* (51). DVOŘÁK deals with the question of the types of dermatophytes (22, 25), 1952 and 1955.

In addition to the above we must touch on the works concerning the causes of exceptionally found types of dermatomycoses. FINGERLAND et soc. (30), 1953, describe the case of a generalized cryptococcosis. FRÁGNER (36), 1955, deals with the incidence of scopulariopsis var. hominis in pathological material and further, FRÁGNER & SVÁTEK (37, 38), 1955—56 deal with the incidence of *Sporotrichum Gougeroti* and *Sporotrichum Schenckii* in the spaces between miners' toes. ZDRÁHAL et soc. (92), 1956, deal with the nocardiosis of the lungs, the brain and brain membranes.

References

1. Balun, E., Microcultures of dermatophytes, Čsl. Derm. **31,** 93, 1956.
2. Benca, J., Treatment of deep trichophytosis with podophyllin, Čsl. Derm. **29,** 202, 1954.
3. Blatnická, B., Therapy of dermatomycoses at the Klinic of Košice, Čsl. Derm., **31,** 149, 1956.
4. Buchníček, J., Influence of chloromycetone on the breathing of Trichophyton gypseum, Čsl. Derm. **31,** 311, 1956.
5. Chmel, L., The incidence of dermatophytes in Slovakia during the period 1942—1946, Bratislavské lekárs. Listy **26,** 601, 1946.
6. Chmel, L., About the epidemiology and prevention of dermatomycoses, Slov. Lek. **9,** 1, 1947.
7. Chmel, L., Mycotic diseases of the skin in relation to military service, Voj. zdravotn. Listy **17,** 333, 1948.
8. Chmel, L., The problem of epidermophytosis in sport, Slov. Lek. **11,** 347, 1949.
9. Chmel, L., Nižnánsky, F. & Oravec, C., Survey of dermatomycoses in Slovakia, Slov. Lek. **12,** 431, 1950.
10. Chmel, L., Some actual problems of fight against dermatomycoses in Czechoslovakia, Lek. Obzor **2,** 381, 1953.
11. Chmel, L. & Babák, A., Clinical and mycological study of superficial trichophytosis provoked by the strain Trichophyton sulfureum, Čsl. Derm. **29,** 189, 1954.
12. Chmel, L. & Pálešová, K., Contribution to the epidemiology of favus in Slovakia, Čsl. Derm. **29,** 196, 1954.
13. Chmel, L. & Pálešová, K., A mycological study of the morbific agent of favus, cultivated at our Clinic, Bratislavské lekárs. Listy **34,** 991, 1954.
14. Chmel, L. & Hegyi, E., Notes on the organization of fight against dermatomycoses in the country. Lek. Obzor **4,** 681, 1955.
15. Chmel, L., The problems of epidemiology and fight against trichophytosis, microsporosis and favus, Bratislavské lekárs. Listy **36-II,** 641, 1956.
16. Chmel, L., An epidemiologic study about the incidence of a new dermatophyte in our mykoflora — Trichophyton rosaceum, Bratislavské lekárs. Listy **36-II,** 656, 1956.
17. Chmel, L. & Pálešová, K., Some experiences from a field study of dermatomycoses in agriculture, Bratislavské lekárs. Listy **36-II,** 669, 1956.
18. Chmel, L. & Kuželová, K., A comparative study of the effectiveness of some of our disinfecting preparations on dermatophytes, Bratislavské lekárs. Listy **36-II,** 684, 1956.
19. Chmel, L., Epidemiology and prevention of trichophytosis, microsporia and favus, Plísňové choroby kožní, Státní zdravotnické nakladatelství, Phraha, 1956.
20. Chmel, L., An epidemiologic analysis of the situation of dermatomycoses in agriculture (statistical part), Bratislavské lekárs. Listy **37-I,** 73, 1937.
21. Chmel, L., Idem (epidemiological part), Bratislavské lekárs. Listy **37-I,** 140, 1957.
22. Dvořák, J., Kinds of dermatophytes, Voj. zdravotn. Listy, **21,** 340, 1952.
23. Dvořák, J., Patočková, M., Dočekal, M. & Dvořáková, D., Results of the examination of dermatomycoses, Sborník prací VLA II, 120, 1953.
24. Dvořák, J. & Dočekal, M., Treatment of dermatomycoses with formic acid, Voj. zdravotn. Listy No. 6, 1953.
25. Dvořák, J., The question of genus in the case of dermatophytes, Sbor. Prací (Voj. lék. Akad.) III, 60, 1955.
26. Dvořák, J., New interdigital mycosis? Sbor. Prací (VLA), IV, 97, 1956.
27. Dvořák, J. & Výmola, F., Instructions for the prophylaxis of candidoses in antibiotic treatment, Lékařské zprávy VLA, No. 9, 109, 1956.
28. Erbák, V., Trichophytosis caused by zoophilic strains, Lek. Obzor **4,** 600, 1955.

29. Fabian, A., Our experience with the treatment of epidemics of microsporosis, Čsl. Derm. **31**, 105, 1956.
30. Fingerland, A., Vortel, V., Dvořák, J. & Zdráhal, F., Generalized cryptococosis, Čas. Lék. čes., No. 30, 1953.
31. Frágner, P. & Horák, V., Mycostatic effect of isofurancarbonic acid. Čsl. Derm. **30**, 230, 1955.
32. Fágner, P. & Krauskopf, J., Trichophyton Schönleini Lebert 1843 var. album Sabouraud 1908, the origin of epidemics and epizootics in agriculture, Čes. Mykol., **9**, 165, 1955.
33. Frágner, P. & Krauskopf, J., Antimycotic effect of chinosol, Čsl. Derm. **31**, 15, 1956.
34. Frágner, P. & Kvita, V., A contribution to the mycostatic effect of some derivates of chinolin, Čsl. Derm. **31**, 220, 1956.
35. Frágner, P. & Krauskopf, J., Trichophyton gypseum Bodin 1902 var. Quinckeanum Quincke 1885, Blanchard 1896 the cause of epidemics in the treatment of straw, Čsl. Hyg., Epid. Mikrobiol. **5**, 47, 1956.
36. Frágner, P., Scopulariopsis var. hominis Brumpt et Langeron 1910 in our pathologic material, Čsl. Derm. **30**, 287, 1955.
37. Frágner, P. & Svátek, J., Sporotrichum Gougeroti Matruchot in our material Čes. Mykol., **9**, 139, 1955.
38. Frágner, P. & Svátek, J., Sporotrichum Schenckii Matruchot in our material, Čes. Mykol., **10**, 217, 1956.
39. Frágner, P., Candida albicans (Robin 1853) Berkhout 1923, some current cultivated forms, Čes. Myckol. **10**, 251, 1956.
40. Godovič, A., Problematics of increased incidence of trichophytosis in East Slovakia from the standpoint of the dermato-venerologic section of the Michalovce hospital and material collected there. Čsl. Derm. **31**, 205, 1956.
41. Groh, J., Use of penicillin and streptomycin in the isolation of dermatophytes, Čsl. Derm. **29**, 187, 1951.
42. Groh, J., The incidence of dermatophytes in the region of Hradec Králové, Čsl. Derm. **31**, 152, 1956.
43. Hegyi, E., On the question of hypersensitiveness to Trichophytin, Lék. Listy **9**, 109, 1954.
44. Hegyi E., Kuželová, K. & Lendvai, O., Incidence of professional epidermophytosis in work with bristles, Bratislavské lekárs. Listy **36-II**, 677, 1956.
45. Hegyi, E., Fight against epidermophytoses and superficial blastomycoses in factories, Plísňové choroby kože, Stát. zdrav. nakl., Praha, 1956.
46. Hegyi, E., Chmel, L. & Martin, K., New arrangement for luminiscent examination adapted to field conditions. Bratislavské lekárs. Listy **37-I**, 304, 1957.
47. Hejtmánek, M., Unusual forms of variability in Trichophyton mentagrophytes, Fol. biol. **II.**, 159, 1956.
48. Hejtmánek, M., Electron microscopy of dermatophytes, Scripta med. **29**, 13, 1956.
49. Hejtmánek, M., Transformation of aerial lanuginous colonies of Trichophyton Kaufmann-Wolf into faviforme colonies, Čsl. Biol. **5**, 171 1956.
50. Hejtmánek, M., Development of polymorphous colonies Trichophyton Kaufmann-Wolf, Čsl. Biol. **5**, 163, 1956.
51. Hejtmánek, M., Presence of coremia in cultures of Trichophyton Kaufmann-Wolf, Čes. Mykol. **9**, 9, 1955.
52. Hejtmánek, M., The stimulating effect of certain conditions on the development of coremiae, Scripta med., **29**, 101, 1956.
53. Hejtmánková, N., Polymorphy of the dermatophyte Tr. Kaufmann- Wolf and some of its laws, Scripta med., **28**, 208, 1955.
54. Horáček, J., Our experience with the treatment of Sycosis barbae mycotic, Prakt. Lék. **26**, 305, 1946.
55. Horáček, J. & Polster, M., Fungicidal effect of phenol derivates, Dermatologica (Basel) **96**, 342, 1948.
56. Horáček, J. & Langer, J., Mass infection of school children caused by Trichophyton sulfureum, Čsl. Derm. **31**, 107, 1956.

57. Hübschmann, K., Langerová, M. & Langer, J., An attempt at determining a complex microbial picture of the skin, Čsl. Derm. **29**, 103, 1954.
58. Hübschmann, K., Krauskopf, J. & Frágner, P., Trichophytic diseases in agriculture, Čsl. Derm. **31**, 7, 1956.
59. Hübschmann, K., Krauskopf, J. & Frágner, P., A contribution to the explantation of the incidence of mycoses in the 3rd and 4th interdigital space of the right hand, Čsl. Derm. **31**, 47, 1956.
60. Hübschmann, K., Krauskopf, J. & Frágner, P., A contribution to the determination of the average picture of the mycotic flora of the skin, Čsl. Derm. **31**, 211, 1956.
61. Hübschmann, K. & Frágner, P., Interdigital erosions in pickers of goose-feathers, Čsl. Derm. **30**, 195, 1955.
62. Janda, J., Principles of prevention of mycotic diseases of the skin, Čsl. Derm. **29**, 110, 1954.
63. Janda, J., Principles of prevention of mycotic diseases transfered from animals to man, Plísňové choroby kožní, Stát. zdrav. nakl., Praha, 1956.
64. Kabelík, J., Fungicida, Čsl. Derm. **29**, 161, 1954.
65. Kachnič, M., Mycotic diseases at the dermatological clinic in Košice in the years 1946—1954, Čsl. Derm. **31**, 141, 1956.
66. Langer, J., A contribution to the standardization of trichophytine, Čsl. Derm. **25**, 293, 1950.
67. Langer, J., Microsporum ferrugineum — its first isolation on the territory of Czechoslovakia, Čsl. Derm. **28**, 446, 1953.
68. Liška, J., The reactive component of trichophytine, Čsl. Derm. **31**, 276, 1956.
69. Neumann, E., Hyaluronidase in the therapy of onychomycoses, Čsl. Derm. **31**, 12, 1956.
70. Nižnánsky, F., Survey of dermatomycoses in Slovakia, Čas. čsl. Veter. 324, 1950.
71. Nižnánsky, F., Fight against trichophytosis of cattle, Veterinářství 82, 1952.
72. Nižnánsky, F., Fight against rtichophytosis in Czechoslovakia, Veterinářství 204, 1953.
73. Nosek, J. & Dvořák, J., The disinfection of mycose-infected equipment with ethylenoxyde. Voj. zdravotn. Listy **23**, No. 11, 1954.
74. Obrtel, J., Differential clinical diagnosis and therapy of oidio- and dermatophyto-onychoses, Čsl. Derm. **24**, 217, 1949.
75. Obrtel, J., "Id's" in dermatology, Čsl. Derm. **25**, 83, 1950.
76. Obručník, M., On the changes in the normal histological structure of the skin in the interdigital spaces on human feet and their relation to the pathogenesis of the skin diseases found in our countries, Čsl. Derm. **28**, 112, 1953.
77. Pálešová, K., Study of the incidence of favus in the region of Bratislava, Bratislavské lekárs. Listy **33**, 577, 1953.
78. Polster, M. & Horáček, J., Fungicidity of phenol derivates, Čsl. Derm. **26**, 371, 1951.
79. Polster, M., Mechanics of the antimycotic effect of the derivates of paranitrophenol **I**, Čsl. Derm. **29**, 184, 1954.
80. Sedláček, V., Evaluation of the experimental knowledge gained in the work with home made trichophytine, Čsl. Derm. **25**, 3, 1950.
81. Sedláček, V., Diagnosis, therapy and prevention of dermatomycoses most often met with in praxis. Voj. zdravotn. Listy **19**, 74, 1950.
82. Sedláček, V., Candida as cause of balanoposthitis, Čsl. Derm. **26**, 64, 1951.
83. Sedláček, V., Results of mycologic examination at the skin Clinic of Brno in the years 1946—1951, Lék. Listy **7**, 106, 1952.
84. Sedláček, V., Treatment of trichophytosis with CO_2 snow, Lék. Listy **7**, 552, 1952.
85. Sedláček. V., Antimycotic properties of plant siliae and of some other substances in vitro, Čsl. Derm. **29**, 175, 1954.
86. Sedláček, V., Microsporon ferrugineum in children's groups, Čsl. Pediatr. **10**, 756, 1955.
87. Štáva, Z., Remark on the work of E. Neumann "Hyaluronidase in the therapy of onychomycoses", Čsl. Derm. **31**, 13, 1956.

88. Štekláč, B., Drgonec, J. & Ružička, A., A contribution to the problematics of dermatomycoses, Voj. zdravotn. Listy **22**, 257, 1953.
89. Uličná-Autratová, L., The effects of chlornitrophenol on Candida albicans and on Achorion Schönleini in vitro, Čsl. Derm. **31**, 87, 1956.
90. Vacátko, S., Favus squamosus, Čsl. Derm. **31**, 19, 1956.
91. Viščor, A., Survey of dermatophytes in agricultural workers and in those engaged in animal production, Bratislavské lékars. Listy **35**, 265, 1955.
92. Zdráhal, F., Beran, J., Fingerland, A., Vortel, V., Herout, F. & Dvořák, J., Nocardiosis of the lungs, the brain and brain membranes, Sbor. Prací (Voj. lék. Akad.) **4**, 97, 1956.

LIBRI NOVI

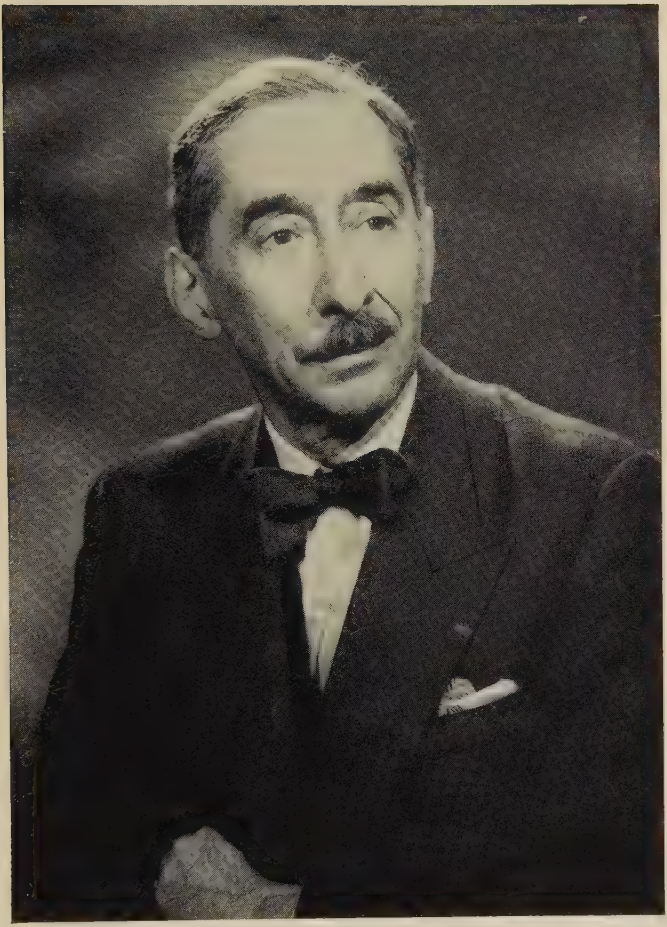
Allergie und Allergische Erkrankungen, edited by E. RAJKA, with the collaboration of R. BACKHAUS, K. FARKAS, I. HUSZAK, M. JULESZ, L. KESZTYÜS, L. MOSONYI, E. RAJKA, I. WENT and E. WINKLER. — With 74 illustrations and 18 tables. Vol. I. 638 pp. Publishing House of the Hungarian Academy of Sciences, Alkotmány u. 21. Budapest, V. (German)

This volume I of the Handbook on Allergy and Allergic Diseases (in 2 volumes) presents the general theoretical concepts of Allergology with its close correlation with immunology, with chapters on immunochemistry, serology and biochemistry of neuro-allergic processes. It contains 14 chapters. Every chapter attests to the great erudition of its author and thorough knowledge of his subject. Every chapter has a detailed bibliography. This reviewer regrets very much that these references are "blind", they do not contain the titles of the papers. This fact detracts a great deal of the usefulness of such references. The illustrations are well chosen and well reproduced. The tables are well organized. The original of this work was written in Hungarian, this is a translation by O. RÁTZ and M. CUKOR. The translation is excellent, fluent and well readable. The production of the book is most satisfactory. Despite of a voluminous literature on Allergology, handbooks included, this work is a welcome addition in the family. It attests to the high standard of Hungarian research and erudition. It can be highly recommended to all those with genuine interest in basic sciences, among them, also to mycologists.

TIBOR BENEDEK

ERRATUM

Vol. XI, page 250 and plate I: the year of Professor TRAVERSO's death should read: 1955.

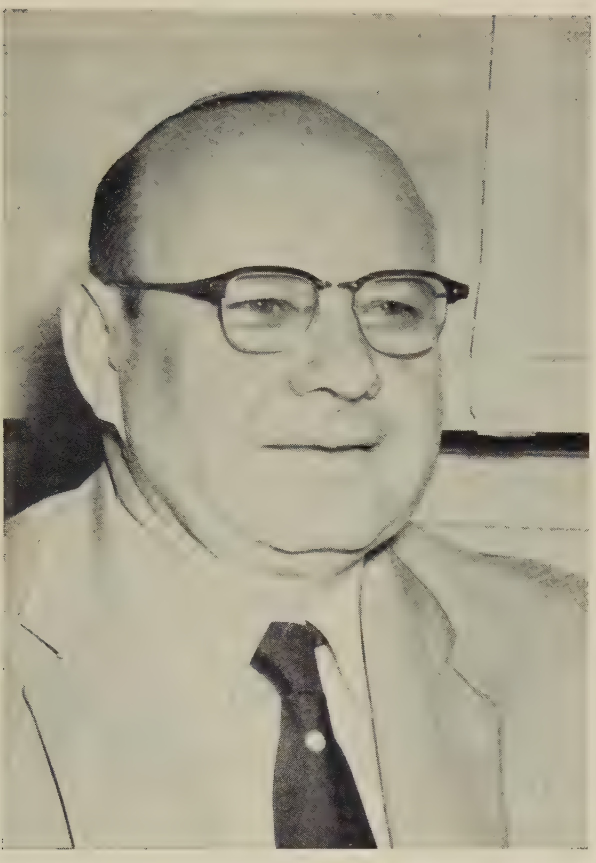


E. RIVALIER

Chef de Laboratoire Hôpital Saint-Louis
Paris, France

Gallery of Contemporary Noted Mycologists

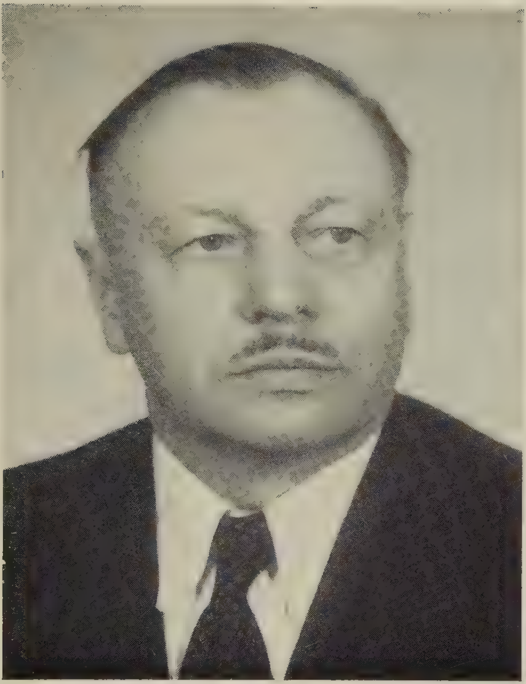
61



EMIL MRAK
Department of Food Technology
University of California
Davis, Calif., U.S.A.

Gallery of Contemporary Noted Mycologists

62



K. CEJP

Department of Mycology and Plant Pathology
Charles University
Praha, C.S.R.

Gallery of Contemporary Noted Mycologists

63

P. PINETTI

Clinica Dermatologica e Centro Micologico
Università di Cagliari, Italia

Gallery of Contemporary Noted Mycologists

64

PRODUCTION OF CRYSTALS BY ROZITES GONGLYOPHORA

by

WALTER BORZANI

Department of Chemistry, Escola Politécnica, University of São Paulo,
São Paulo, Brasil.

(22.IX.1959)

Several species of tropical and subtropical ants have the habit of cultivating and feeding upon fungi. The fungi grow in underground nests, in culture medium prepared by the ants from plant tissues, and produced bromatia, swollen, roundish hyphal tips, which are consumed by the ants (F. A. WOLF & F. T. WOLF, 1949, The fungi, second printing, John Wiley & Sons, Inc., London). Experiments are in progress in our laboratory in order to verify the activity of *Rozites gonglyophora*, a fungus cultivated by *Atta sexdens rubropilosa,* as a probable antibiotics producer.

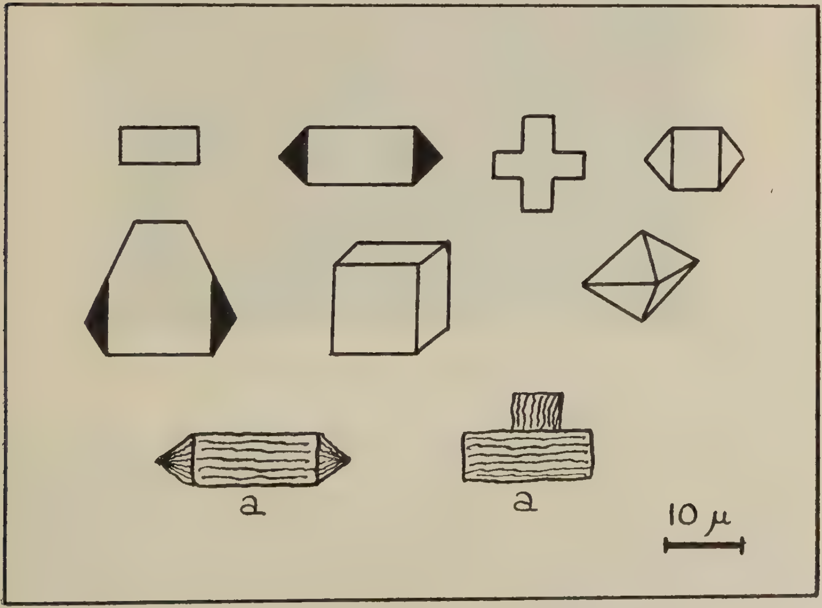


Figure 1. Types of crystals produced by *Rozites gonglyophora.*
(a) Crystals deformed by heating.

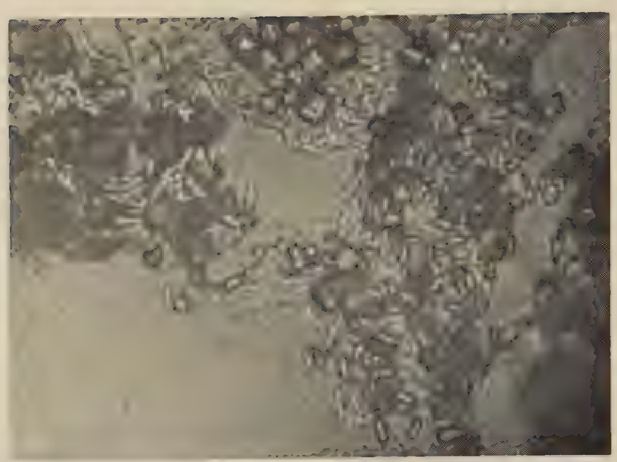


Figure 2. Agglomerate of crystals and dry mycelium.
1 mm = 5 μ

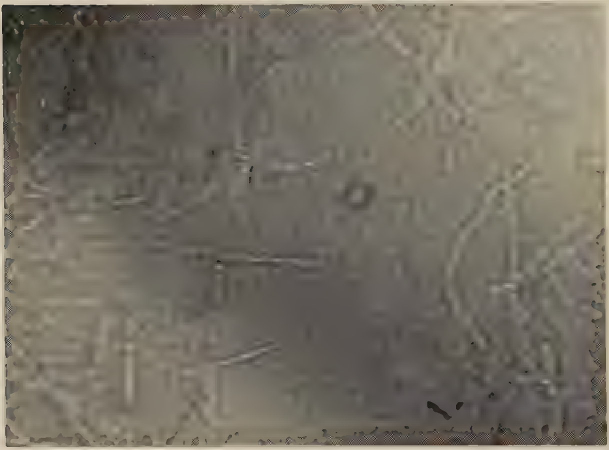


Figure 3. Hyphae of *Rozites gonglyophora* and some isolate crystals.
1 mm = 5 μ

A culture of *R. gonglyophora* was obtained from the artificial nests of *Atta sexdens* of the Instituto Biológico de São Paulo, and was cultivated in glucose agar and in banana solid medium (BORZANI & PULLIN, Ciência e Cultura, 6(1), 21, 1954). Some cultures were also made in glucose broth, with and without aeration. In all the cases studied, the fungus produced large quantities of crystals that remain between the hyphae. Figure 1 shows the aspects of the observed crystals. Figures 2, 3 and 4 are photographs of some preparations.

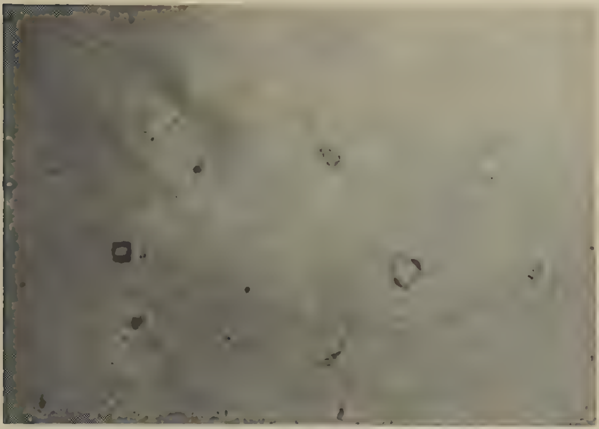


Figure 4. Isolate crystals produced by *Rozites gonglyophora.*
1 mm = 5 μ

Figure 5. Crystals heated at 350° C.
1 mm = 5 μ

Temperature as high as 350° C only produces small crystal deformations (Figures 1, 5 and 6).

The crystals are insoluble in water, ethyl alcohol, acetic acid, carbon tetrachloride, dioxane, acetone, chloroform, amylacetate, ammonium hydroxide, but reacts with the following materials: 0.1 to 4.0 N sodium hydroxide; 85 % phosphoric acid; 0.6 to 20 % hydrochloric acid; 1.0 to 34 % nitric acid; 1.5 to 50 % sulfuric acid. A detailed report on this will be presented shortly.

The author wishes to acknowledge the technical assistance of MARINA L. R. VAIRO, and wishes to express his appreciation to Professor ALCEU FABIO BARBOSA for providing the photographs.

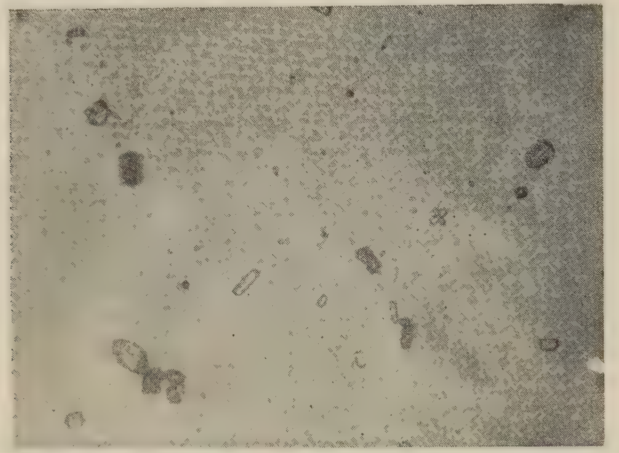


Figure 6. Crystals heated at 350 ° C.
1 mm = 5 μ

PULMONARY MUCORMYCOSIS IN A HARP SEAL CAUSED BY MUCOR PUSILLUS

by

WILLIAM KAPLAN, D. V. M.¹), LEONARD J. GOSS, D. V. M.²),
LIBERO AJELLO, PH. D.¹) & M. SUE IVENS, B. S.¹)

(1.IX.1959)

Mucormycosis, a disease caused by fungi belonging to the class Phycomycetes, usually of the order Mucorales, has been reported in both man (BAUER, et al., 1955, BAKER, 1957) and lower animals (AINSWORTH & AUSTWICK, 1959). The organisms classified in this botanical category are widely disseminated in nature and are not usually considered to be animal pathogens. They are found in soil, water, food and decomposing organic matter where they exist in a free-living state. Although man and lower animals are frequently exposed to these organisms in nature, phycomycete infections are encountered infrequently. In humans, diabetes mellitus, leukemia and other debiliating diseases (BAUER, et al., 1956, and BAKER, 1956) are conditions which have been known to predispose to the development of mucormycosis. In lower animals, however, the association of natural infection with an altered physiological state has not been demonstrated.

To date, several species in the genera *Mucor*, *Rhizopus* and *Absidia* have been incriminated in spontaneous mucormycosis in animals. Members of these three genera are commonly encountered in the laboratory as contaminants. Thus, the mere recovery of phycomycetes from clinical materials does not furnish sufficient proof of their incrimination in a disease process. To establish a diagnosis of mucormycosis, it is essential to demonstrate tissue invasion in addition to recovery of the causal agent. In tissues, members of the genera *Mucor*, *Rhizopus*, and *Absidia* develop broad (6—15 μ) branched, non-septate mycelium. In culture they are characterized by the formation of similar mycelium and production of asexual sporangiospores and, in some cases, by the development of sexual spores that are termed zygospores.

---

¹) From the Communicable Disease Center, Public Health Service, U. S. Department of Health, Education, and Welfare, Atlanta, Georgia.

²) New York Zoological Park, Bronx, New York.

To date, among animals, mucormycosis has been described in cattle, pigs, horses, dogs, mink, guinea pigs, and mice (AINSWORTH & AUSTWICK, 1959).

The present paper describes a case of spontaneous mucormycosis in a harp seal (*Phoca groenlandica*) caused by *M. pusillus*. This is the first authenticated case of mucormycosis in a seal. In addition, the report records the pathogenicity of the causal agent for rabbits rendered diabetic by alloxan.

CASE REPORT

The animal in question, a female harp seal, was captured as an infant in April, 1955 on the ice off the Magdalen Islands, Province of Quebec, Canada. These islands are located north of Nova Scotia, in the Gulf of St. Lawrence. The seal was held captive in a small outdoor pen in the Magdalen Islands, until mid-summer, 1955. Food during this time consisted entirely of locally caught fresh herring. From mid-summer until December, 1955, the animal was housed under cover at the Grand River Biological Station, Grand River, Province of Quebec, where it was fed locally caught fresh fish, frozen herring and mackerel. In December of that year, the seal was shipped by rail express to the Biological Station, St. Andrews, New Brunswick and held during January and part of February in an outdoor tide pool. The diet at this time consisted of fresh and frozen haddock, herring and winter flounder. To supplement its diet during infancy, the seal was fed vitamin pills.

In February, 1956, the harp seal was donated by the Fisheries Research Board of Canada to the New York Zoological Society. Upon arrival at the Bronx Zoological park on February 13, 1956 the animal was placed in a fresh water pool together with two other seals, a female Gray Seal (*Halichaerus gryphus*) and a female European Harbor Seal (*Phoca vitulina*). The food at this time consisted of frozen mackeral. Fresh water was constantly allowed to enter the pool and the excess drained into a sewer. Once weekly, the pool was drained, cleaned and refilled. On March 3, 1958, the harp seal suddenly went off feed and appeared languid. The attending veterinarian, one of the authors (L.J.G.), administered intramuscularly 4 cc of an antibiotic suspension containing 800,000 units of procaine penicillin G and 1.0 g of dihydrostreptomycin sulfate. During the night of March 4, 1956, the seal died. Figure I shows a mature harp seal and her pup.

Pathologic Findings

At autopsy, the body was found to be well nourished. With the exception of the lungs, the organs showed no grossly visible pathology. The lungs were pneumonic and presented a uniformly dark red, "meaty" appearance. On the surface of one lung the attending veterinarian observed a raised, blood-red nodule, 2 cm in diameter. The nodule was not encapsulated and could be readily felt on palpation. A portion of the nodule and adjacent tissue was removed and fixed in 10 % formalin for histological examination. Cultures were prepared with Sabouraud dextrose agar. The clinical diagnosis of the cause of death was pneumonia of undetermined origin.

Fig. 1. A mature female harp seal (*Phoca groenlandica*) and her pup. (Courtesy of Dr. Per Host, Oslo, Norway.)

Histopathologic study of hematoxylin and eosin stained sections disclosed various sized, well delineated granulomata. These had small central foci of liquifaction necrosis immediately surrounded by an area of coagulation necrosis. The outer area in turn was surrounded by mononuclear cells, predominately plasma cells and lymphocytes with a lesser amount of eosinophils and macrophages. Scattered among the granulomata were giant cells, with peripherally aligned nuclei. Within macrophages and giant cells and scattered extracellularly among the granulomata, numerous segments of broad, (6—8 μ) branched aseptate hyphae were noted (Fig. 2). The lung sections also exhibited profuse erythrocytic extravasation into the alveoli. Marginating the granulomatous area the parenchyma contained foci of plasmocytic and lymphocytic infiltration.

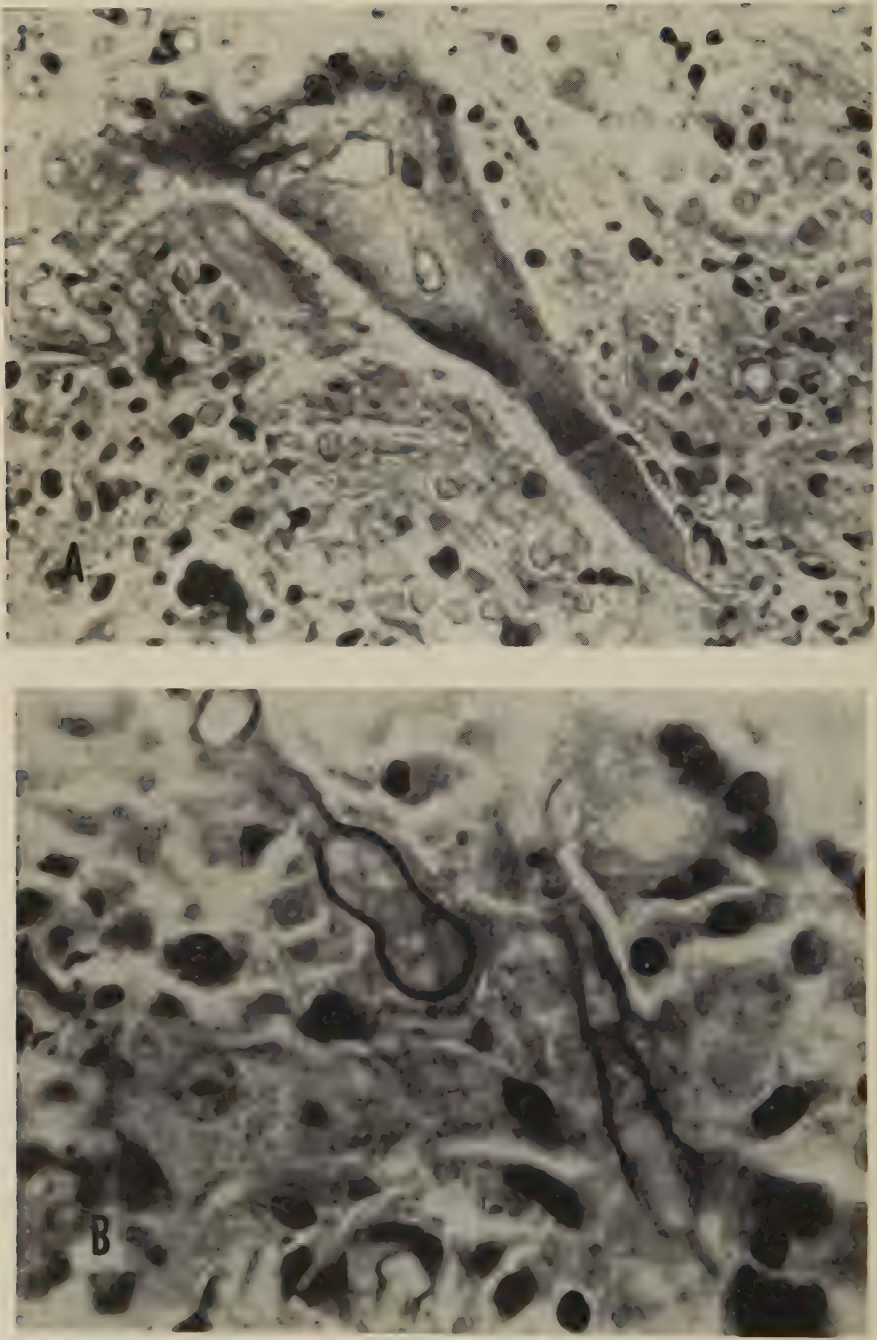


Fig. 2. Hyphae of *Mucor pusillus* in pulmonary granuloma of a harp seal.
A. A giant cell containing hyphae of *M. pusillus*. × 610.
B. Hyphae of *M. pusillus* found extracellularly in granuloma. × 980.

In these areas the alveolar septa were collapsed. Foci of fibrosis of the parenchyma were also seen, surrounding portions of the granulomatous tissue. There was no apparent invasion of blood vessels by the hyphae. With the stain employed, bacteria were not revealed in the affected tissue. On the basis of histopathological findings the infectious process was diagnosed as mucormycosis.

Mycological Findings

The Sabouraud dextrose agar cultures prepared from the seal's lung at autopsy yielded in a fungus which was identified as a member of the genus *Mucor*. The organism was a rapidly growing mold with abundant fluffy aerial mycelium that was at first white, but later became grayish brown in color (Fig. 3-A). Microscopic examination showed branched, nonseptate mycelium. Branched sporangiophores arose singly from the mycelium. The terminal sporangia at maturity contained numerous round sporangiospores (Fig. 3-B). Subcultures were sent to Dr. C. W. HESSELTINE, U. S. Department of Agriculture, Peoria, Illinois, who identified the fungus as *Mucor pusillus*.

EXPERIMENTAL ANIMAL INFECTIONS

An attempt was made to reproduce the disease in laboratory animals with the strain of *M. pusillus* isolated from the seal. For this purpose it was decided to employ rabbits with acute toxic alloxan-induced diabetes (BAUER, et al., 1955 and ELDER & BAKER, 1956). This altered metabolic state was produced in three 1500 g rabbits by a single intravenous injection of a freshly prepared 2 % solution of alloxan in sterile saline. The amount of drug given was 20 mg per 100 g of body weight. Acute toxic diabetes appeared in 72 hours as evidenced by marked acetonuria, glycosuria, excessive thirst, increased excretion of urine, loss of body weight, rapid dehydration and general weakness. At this time the diabetic rabbits were infected by the intranasal instillation of 1.5 ml of a suspension of *M. pusillus* spores in saline. To prevent sneezing, the nostril first was swabbed with a 1 % solution of chloroprocaine hydrochloride. A normal non-diabetic rabbit, that served as a control, was challenged in the same manner. The spore suspension used was standardized to give 35 % light transmittance on a Coleman Universal Spectrophotometer [1]) (Model 14) set at 600 mμ.

The 3 diabetic animals died 48 hours after challenge, at which time they were autopsied and cultures prepared from the lungs and brains. The control rabbit was sacrificed 72 hours after instillation of the spore suspension and cultures also taken from the lungs and brain. At the time of autopsy, impression smears were made from the lungs and brains of all rabbits and stained with methylene blue. Then the brains were fixed in 10 % formalin and portions of the lungs were fixed in Zenker's fluid with 5 % glacial acetic acid. The tissues were then sectioned and stained with hematoxylin and eosin.

---

[1]) Trade names are used for indentification purposes only, and their use does not constitute endorsement of these products by the Public Health Service.

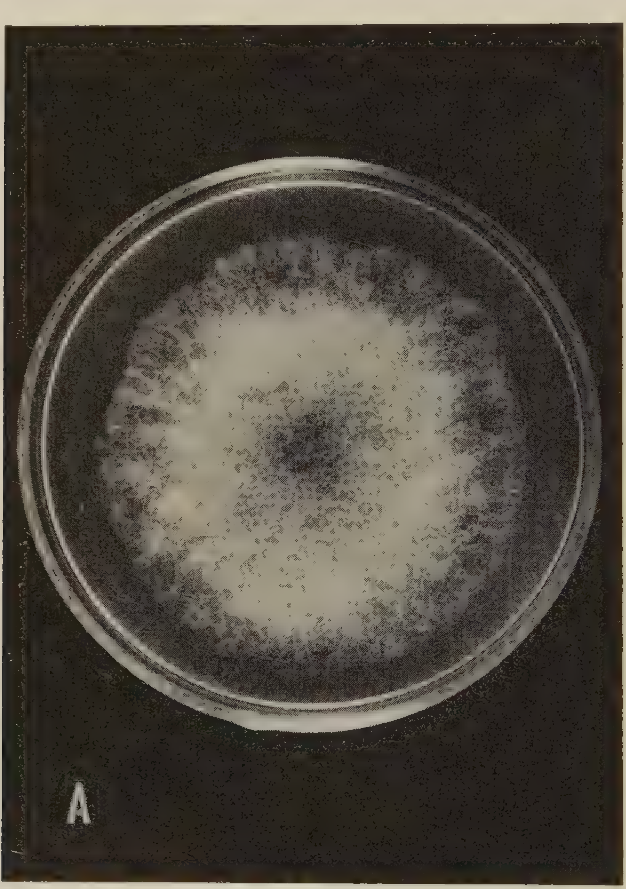
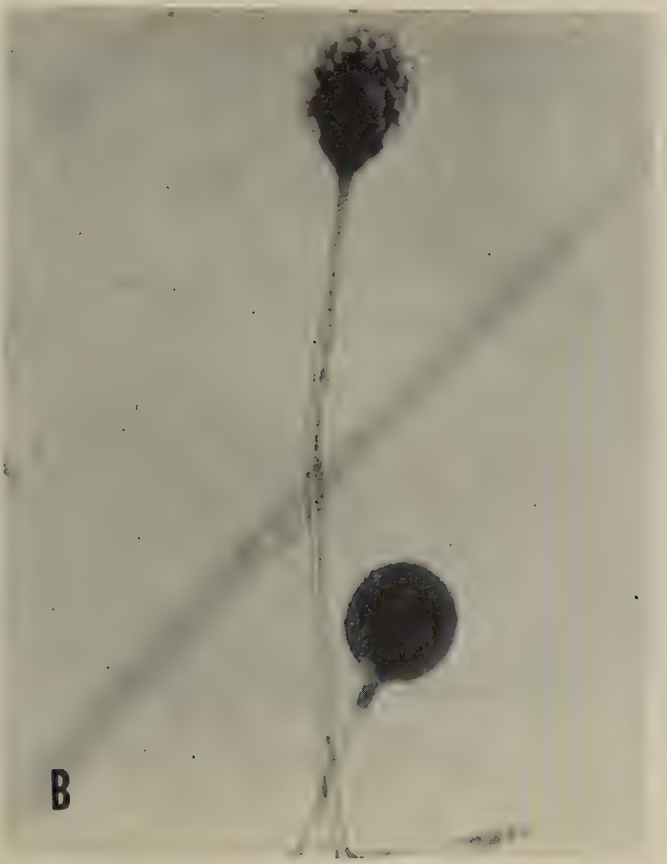


Fig. 3. *Mucor pusillus* isolated from a harp seal.
A. Gross appearance of colony after 7 days cultivation on Sabouraud dextrose agar.

Pathologic Findings

The lungs of the three diabetic rabbits showed dark foci of pneumonia (Fig. 4). Histopathologic study revealed small granulomata with a central area of necrosis. Within the areas of necrosis in some granulomata there was noted the presence of aseptate mycelium (Fig. 6). The necrotic foci were surrounded by many neutrophils and eosinophils and a lesser number of mononuclear phagacytes.

Fungal elements were also found among the inflammatory cells. The granulomata were generally observed adjacent to the large and small branches of the bronchial tree. Within bronchi and bronchioles a dense neutrophilic infiltrate was present. Fungal invasion of blood vessels was not observed.



B. Microscopic appearance of isolate showing a sporangium containing numerous sporangiospores. × 610.

The stained impression smears made from the cut surface of the lungs of two of the infected diabetic rabbits showed branched non-septate hyphae. These findings point to the practicality of employing the impression smear procedure as a rapid diagnostic technique for mucormycosis.

The lungs of the control rabbit showed no gross or microscopic pathology.

The brains of all four rabbits exhibited no significant pathologic changes.

Mycologic Findings

M. pusillus was recovered from the lungs of 1 of the 3 diabetic rabbits. It was not possible to isolate the fungus from the lungs of the control animal nor from the brains of all four rabbits.

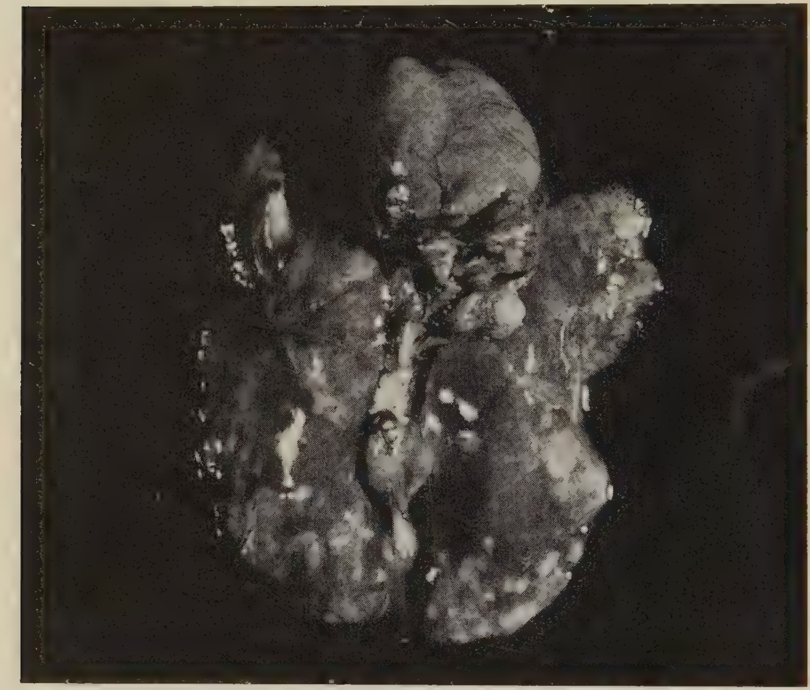


Fig. 4. Dark foci of pneumonia in the lungs of a diabetic rabbit experimentally infected with *M. pusillus*.

DISCUSSION

The present report establishes the susceptibility of aquatic mammals to infection by fungi of the order Mucorales. The causal agent in this case was *M. pusillus*, an organism first described by LINDT in Switzerland in 1886, and since then reported from Germany, Greenland, China and the United States (WOOD, 1955). *M. pusillus* has usually been found to occur in decaying organic matter and soil, where it apparently exists as a saprophyte. To date, spontaneous infections by this fungus have been reported in a horse (TSCHERNIAK, 1928), pig (BAUDET, 1932), calf (AINSWORTH & AUSTWICK, 1955) and, in the present instance, a harp seal. Although its potential pathogenicity for mammals has been established the underlying factors involved in rendering tissues susceptible to infection have not been determined. In man, severe metabolic disorders have been shown to predispose to infection by fungi of the order Mucorcales. The present report records our success in producing experimental infections with *M. pusillus* in alloxan diabetic rabbits. Whether physiological alterations are involved in the pathogenesis of spontaneous mucormycosis in animals remains to be determined. In the case of the harp seal one can only postulate that stress factors generated during transportation to the New York Zoological Park and subsequent exposure to a new environment played a role in the occurrence of this infection. Much remains to be learned concerning the epizootiology of mucormycosis in animals.

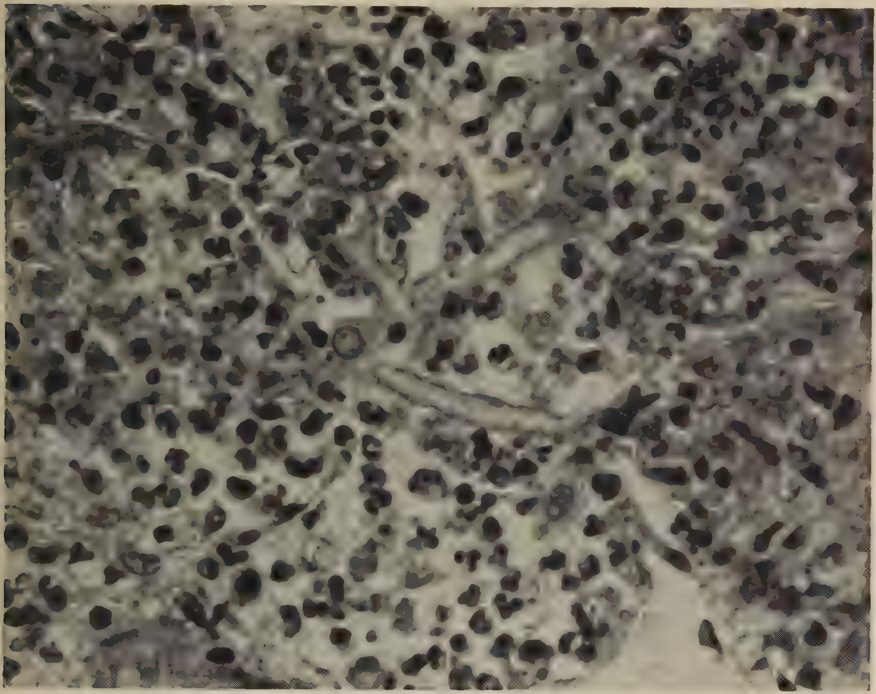


Fig. 5. Hyphae of *M. pusillus* within pulmonary granuloma of a diabetic rabbit. × 980.

Acknowledgment

The authors gratefully acknowledge the guidance of Dr. Heinz Bauer, Department of Pathology, Emory University School of Medicine, Emory University, Georgia in producing the experimental infections in diabetic rabbits. We are also indebted to Drs. WALTER H. SHELDON and HEINZ BAUER, Department of Pathology, Emory University, School of Medicine, for preparation of all stained tissue sections. The authors also express their gratitude to Dr. C. W. HESSELTINE, U. S. Department of Agriculture, Peoria, Illinois for his assistance in the identification of the *M. pusillus* isolate.

References

1. AINSWORTH, G. C. & AUSTWICK, P. K. C. (1955) A survey of animal mycoses in Britain: General aspects. Vet. Rec. **67,** 88—97.
2. AINSWORTH, G. C. & AUSTWICK, P. K. C. (1959) Fungal diseases of animals. Farnham Royal, Bucks, England (Commonwealth Agricultural Bureaux), First Ed., 46—52.
3. BAKER, R. D. (1956) Pulmonary mucormycosis. Amer. J. Path. **32,** 287—313.
4. BAKER, R. D. (1957) Mycormycosis — A new disease. J. Amer. Med. Ass., **163,** 805—808.
5. BAUDET, E. A. R. F. (1932) Mucor pusillus Lindt als oorzaak van mycose bij het varken. Tijdschr. Diergeneesk. **59,** 1163—1164.
6. BAUER, H., AJELLO, L., ADAMS, E. & HERNANDEZ, D. U. (1955) Cerebral mucormycosis: Pathogenesis of the disease. Description of the fungus, Rhizopus oryzae, isolated from a fatal case. Amer. J. Med., **18,** 822—831.
7. BAUER, H., FLANAGAN, J. F. & SHELDON, W. H. (1955) Experimental Cerebral Mucormycosis in rabbits with alloxan diabetes. Yale J. Biol. and Med., **28,** 29—36.
8. BAUER, H., FLANAGAN, J. F. & SHELDON, W. H. (1956) The effects of metabolic alterations on experimental Rhizopus oryzae (mucormycosis) infection. Yale J. Biol. and Med. **29,** 23—32.
9. ELDER, T. D. & BAKER, R. D. (1956) Pulmonary mucormycosis in rabbits with alloxan diabetes. A. M. A. Arch. Path. **61,** 159—168.
10. TSCHERNIAK, W. S. (1928) Zur Lehre von den Broncho- und Pneumonomykosen der Pferde. Arch. Wiss. Prakt. (Tierheilk.), **57,** 417—444.
11. WOOD, J. L. (1955) Mucor pusillus Lindt from Maryland and some notes on its nutrition. Penn. Acad. Sci. **29,** 115—120.

THE MORPHOLOGY OF THE PSEUDOPARASITIC PHASE OF SOME DERMATOPHYTES

by

R. Evron-Maoz & F. Raubitschek, M. D.

From the Mycology laboratory of the Department of Dermatology and Venereology (F. Sagher, M. D., *Director*), *Hadassah University Hospital, Jerusalem, Israel.*

(26.VIII.1959)

In clinical lesions all dermatophytes produce similar hyphal forms which occasionally branch and undergo arthrospore formation. Only on stationary culture do they develop the various complicated fructification organs which allow of their classification. In continuous shake culture, however, certain dermatophytes form only hyphae, arthrospores and chlamydospores. This type of growth has therefore been called the "pseudoparasitic" phase (Raubitschek, 1955).

As the microscopic features of the pseudoparasitic phase do not seem to have been described in detail it was decided to investigate the characteristics of different dermatophytes at intervals during their growth in shake cultures.

Materials and Methods

The fungi used in this study were isolated from patients suffering from various types of tinea, and included 7 strains of *T. mentagrophytes*, 5 of *T. rubrum*, 2 of *T. tonsurans*, 4 of *T. violaceum*, 3 of *T. schoenleini* and 3 of *E. floccosum*. The strains were primarily isolated on brain heart agar with the addition of yeast extract, penicillin and streptomycin, or on potato dextrose agar with the addition of actidion, penicillin and streptomycin. They were subsequently maintained on 2 % glucose agar slants. After attaining full sporulation inocula were prepared by gently scraping the aerial growth from the slant into 8 ml of sterile physiological saline previously added to each tube. After centrifuging and repeated

---

This work has been aided by a grant from the Frances Pascher Research Fund in Dermatology.

washing with sterile saline solution the sediment was resuspended in 2 ml of saline to provide the stock suspension of the organism.

The medium consisted of 0.3 % ammonium sulfate, 0.2 % dibasic potassium phosphate, 0.005 % magnesium sulfate, 0.005 % calcium chloride and 0.01 % soluble yeast extract (Difco) in distilled water. In a few experiments 1 % dextrose was also added (RAUBITSCHEK, 1955). The media were dispensed in 100 ml quantities in 250 ml Erlenmeyer flasks and sterilized for 15 minutes at 15 atm. in the autoclave. 0.2 ml of the stock suspension served as inoculum for each 100 mls of medium. The inoculated flasks were shaken continuously on a Ross-Kershaw machine at 148 strokes per minute for periods of up to 5 weeks. At intervals, ranging from one to four days according to the rate of growth, material was aseptically removed, teased out and examined under the microscope.

RESULTS

General remarks.

There was no essential difference in the morphology of the organisms grown on the medium with and without added dextrose.

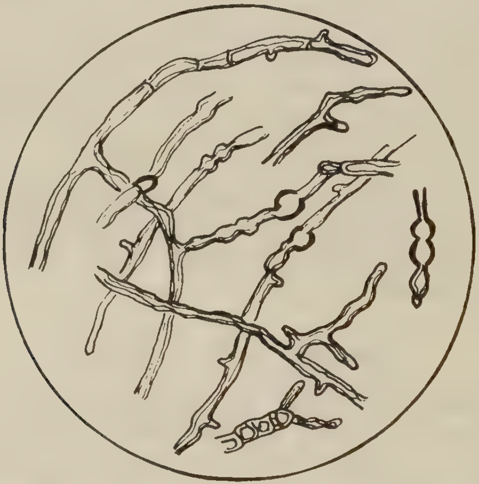


Fig. 1. *T. gypseum.* Hyphae with beginning of arthro- and chlamydospore formation.

In both cases the fungi grew in the form of pellets 2 to 5 mm diameter. However, in the dextrose containing broth the growth was faster and in addition to pellets the organisms grew in the form of a thick band of about 4 mm breadth situated on the wall of the flask at the maximal height reached by the liquid during shaking. This band was covered with aerial mycelium and various fructification organs, according to the organism. As parts of this aerial growth, on maturing, tended to fall off into the medium and

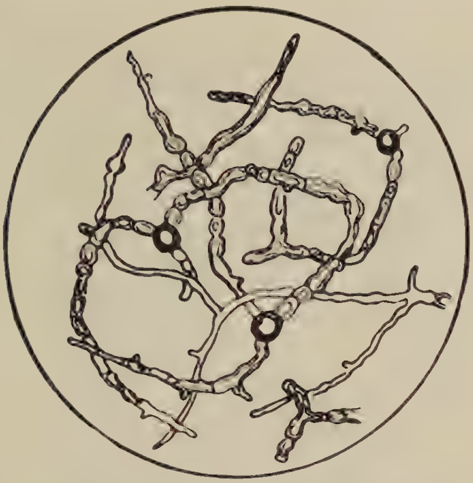


Fig. 2. *T. gypseum.* Arthro- and chlamydospores.

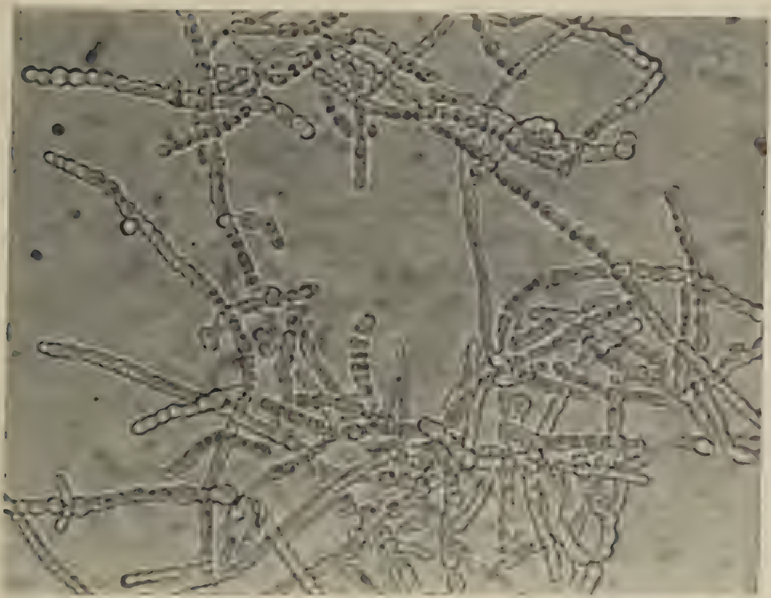


Fig. 2a. Arthrospore formation.

thus confuse the morphological picture of the pelletal growth, the use of sugar containing media was discontinued.

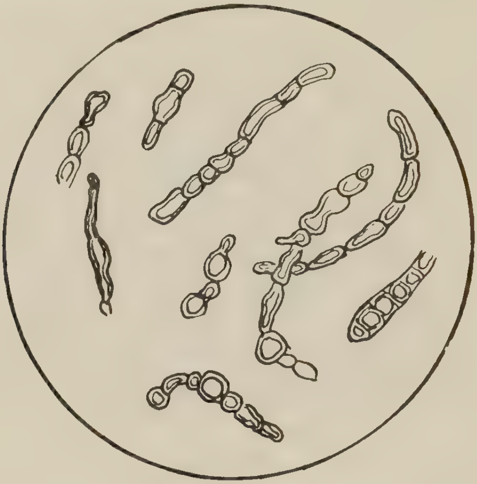


Fig. 3. *T. gypseum.* Arthro- and chlamydospores. Macroconidium - like structure.

T. mentagrophytes.

After 48 hours the growth consisted of branching segmented and non-segmented hyphae with fairly regular and straight contours. Some hyphae showed concentrations of granular dense plasma alternating with empty looking parts of about the same length. In some hyphal walls there were regular roundish swellings giving the filament a monili-form appearance. Later crosswalls were formed at the constrictions and thus the hyphae were transformed into chains of arthrospores. The arthrospores, at first rectangular, later became more and more rounded, matured and broke up into single spores. After two to three days' further growth intercalary or terminal thick walled chlamydospores also appeared. Their number and size increased steadily and by the 8th to 11th day a fairly large number of chlamydospores were present, some even in chains. Many hyphae had by now undergone arthrospore formation and numerous free round or oval arthrospores of about 4 μ diameter were seen. Some hyphae showed terminal, clublike, thick walled plasma-filled swellings. These swellings which were separated from the rest of the hypha by a thick crosswall, were mostly unicellular, but several were divided into two or more compartments, thus forming structures very similar to the macroconidia of the conventional saprophytic phase. The plasma-rich parts of the hyphae had also become thicker, contained one or more crosswalls and were separated from each other by shadowy thinner empty looking parts. Other hyphae were branched and irregularly thickened and the chlamydospores varied in size and form. No further new structures appeared in the later stages but the multiplication of all the above elements produced an increased polymorphism.

T. tonsurans.

After 4 to 5 days regular branching segmented and non-segmented hyphae were seen in some of which there was appreciable arthrospore formation. Numerous chlamydospores were present, mostly intercalary, and concentrations of cytoplasm alternated with empty looking hyphal sections. After 8 days chains of mature arthrospores and a large number of variously sized and formed chlamydospores were present. The plasma-rich parts of the hyphae had increased in thickness, contained one or more crosswalls, and resembled macroconidia of the saprophytic phase. After 11 days no new structures had appeared but those described were more numerous.

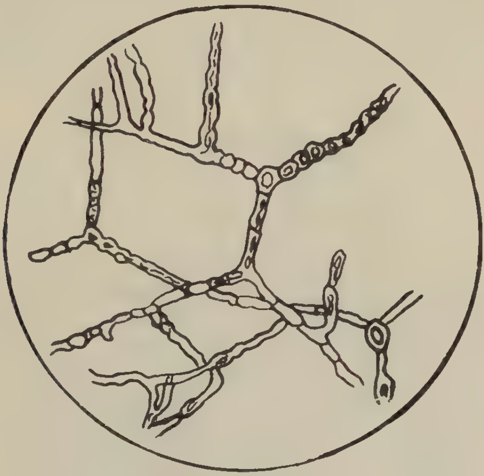


Fig. 4. *T. tonsurans.* Hyphae, beginning arthro- and chlamydospore formation.

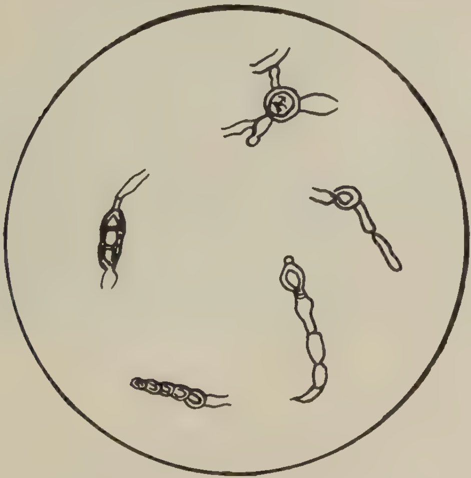


Fig. 5. *T. tonsurans.* Various elements.

T. rubrum.

The growth of this organism was considerably slower than that of the preceding fungi. After 7 days regular, branching, partly segmented hyphae were present, some with monili-form swellings. Parts of the hyphae had begun to increase in thickness and plasma content. No chlamydospores were seen. After ten days some hyphae had become transformed into chains of arthrospores, which were mainly elongated or elliptical and which only became rounder after 2 weeks. At this time chlamydospores were very few and mainly intercalary. Some of the thickened sections of the hyphae had developed into two- or more-celled structures resembling macroconidia of the saprophytic phase. After 4 weeks active growth still continued, but no new structures appeared. A very moderate number of comparatively small chlamydospores was present.

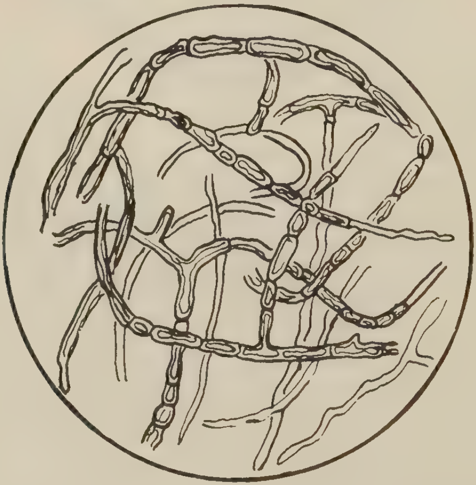


Fig. 6. *T. rubrum.* Hyphae, arthrospores.

T. violaceum.

After 3 days hyphae with straight or wavy contours, 3 to 4 μ thick, could be seen. Some had short lateral knob-like protrusions which represented beginnings of side branching. Some hyphae showed segmentation and others the beginning of arthrospore formation. Chlamydospores had begun to appear. After 8 days the branching had progressed and the hyphae were becoming extremely irregular in outline and thickness. The initially rectangular arthrospores had increased in number and were rounder. Both terminally and intercalary situated chlamydospores were much more numerous, larger and more irregular in shape and size. Some had a diameter of 10 or more micra. They occurred either singly or in clusters and chains, and in much larger numbers than in the preceding organisms.

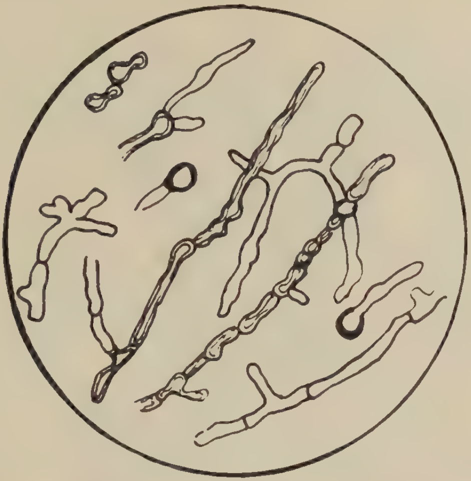


Fig. 7. *T. violaceum.* Hyphae, side branches, arthro- and chlamydospores.

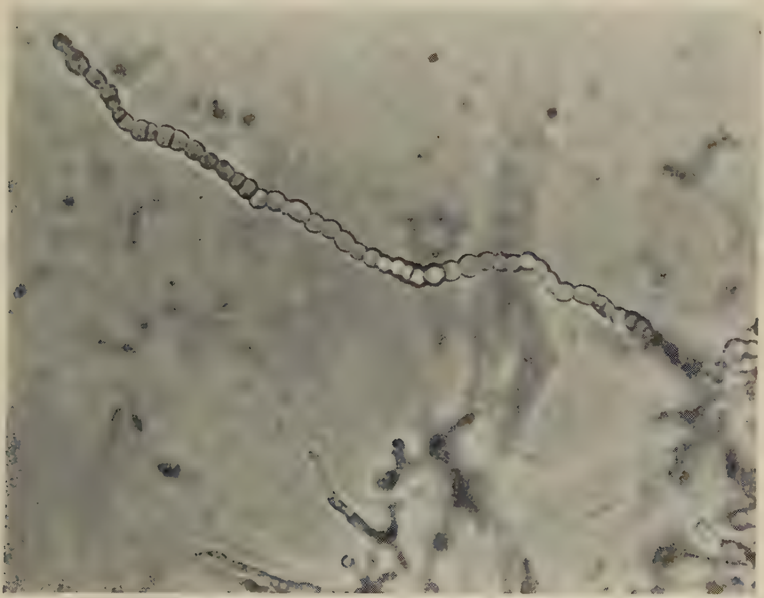


Fig. 7a. *T. violaceum.* Arthrospore formation.

After 3 to 4 weeks the arthrospores became rounded and the number and polymorphism of the chlamydospores and hyphae had further increased. After 35 days the field was almost entirely filled by thickwalled variously sized and shaped chlamydospores, and only a few other structures could be seen.

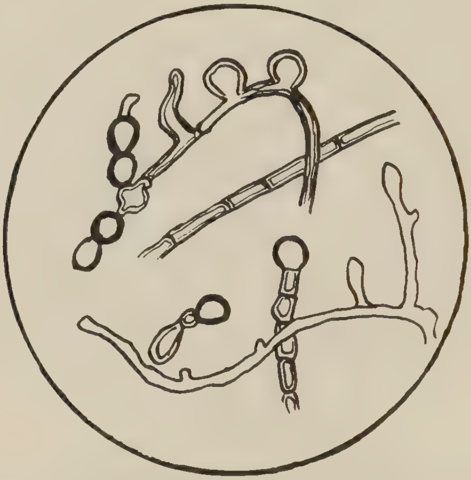
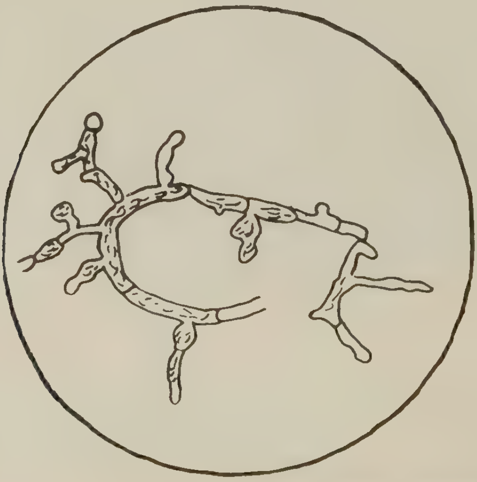


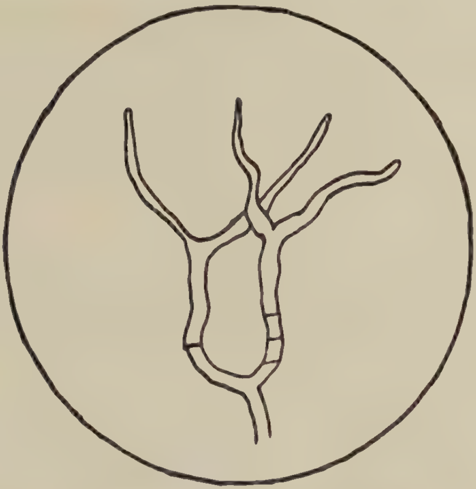
Fig. 8. *T. violaceum.* Chlamydo- and arthrospores.

T. schoenleini.

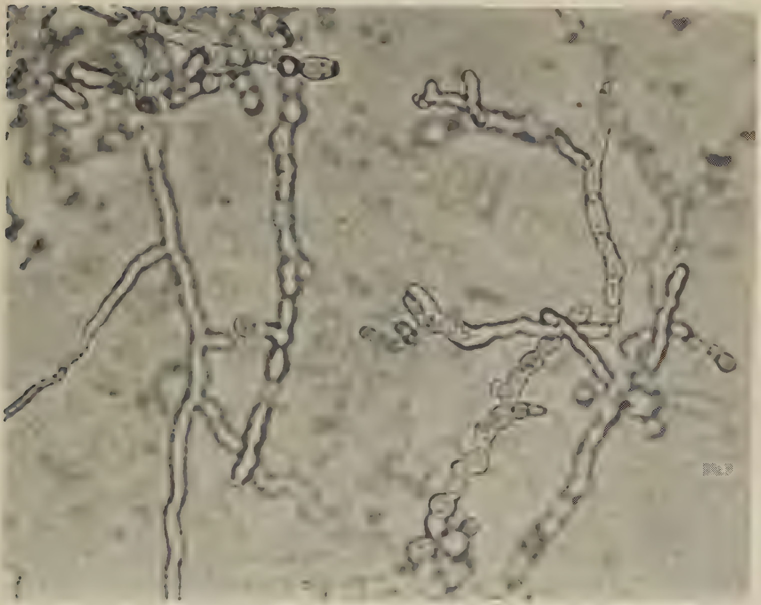
The 6 day growth showed segmented and non-segmented hyphae of very varying thickness, with straight or wavy contours. Some were densely filled with granular yellowish-tinted plasma whereas others seemed empty. In some hyphae plasma-rich and empty looking sections alternated. Some hyphae had lateral or terminal knob-like swellings, and some terminal portions presented two or more short swollen branches, somewhat resembling blunt fork-like



Figs. 9. *T. schoenleini.* Antlers, Pectinate body.

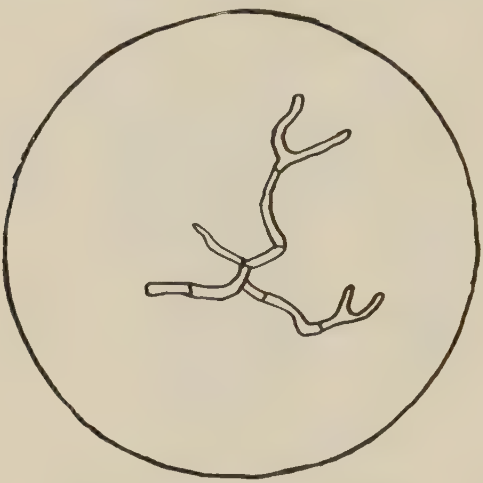


Figs. 10. *T. schoenleini.* Antlers,

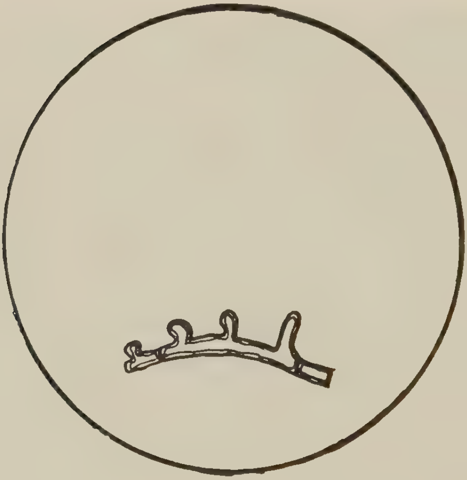


Figs. 11. *T. schoenleini.* Antlers, chandeliers,

structures. A few hyphae showed arthrospore formation the individual spores being unequal in size. A few intercalary and terminal chlamydospores could be seen. The culture in the early phase of its development thus already showed a remarkable measure of



Figs. 12. *T. schoenleini.* Antlers.



Figs. 13. *T. schoenleini.* Pectinate body.

polymorphism. After 10 days irregularity of the hyphae was very marked, there being variously sized and shaped lateral knobs and protuberances, giving a resemblance to knotty and knobby tree trunks. After 2 weeks some of the swollen endings had considerably elongated, or had become almost pointed. This gave a marked resemblance to chandeliers or, where intensive branching had occurred, to complicated antlers. Some hyphae had regularly spaced lateral knobs on one side only, resembling the pectinate bodies of the saprophytic phase. Irregular shaped arthrospore formation continued. The number of chlamydospores increased, as did differences in their form and size. After 4 to 5 weeks no new structures appeared but the increasing variation in the shape and size of the various formations, together with the small number of irregular arthrospores and chlamydospores, produced a marked polymorphous appearance. The number of arthrospores formed was less than with the other organisms.

E. floccosum.

After 3 to 4 days growth there were straight regular hyphae of about 3 to 4 micra thickness. Occasional segmentation and branching could be seen. No arthro- or chlamydospore formation

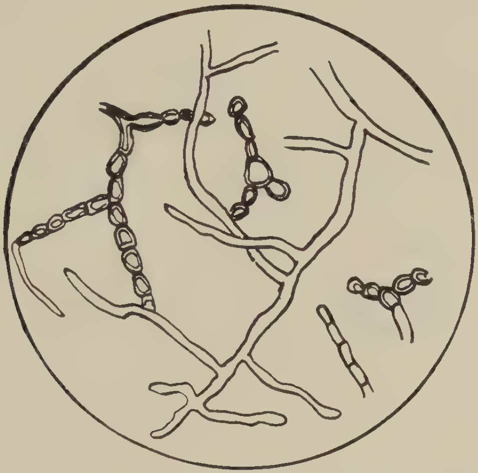
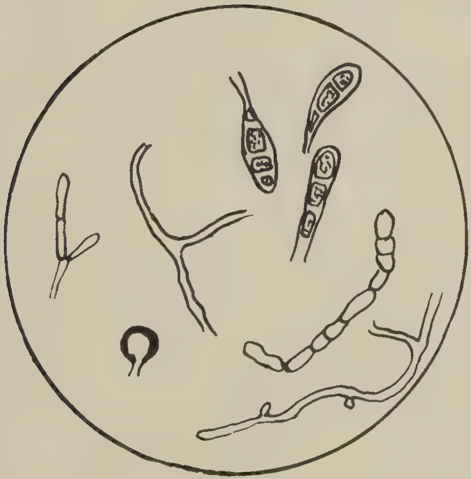


Fig. 14. *E. floccosum.* Arthrospore formation.



Figs. 15. *E. floccosum.* Macroconidia.

was apparent. Some hyphae showed alternate plasma-filled and empty looking sections. After 7 days many hyphae had formed rectangular arthrospores some of which had rounded corners. A few chlamydospores were to be found. After 11 days many chains of rectangular or round arthrospores could be seen and also many variously shaped and sized intercalary or terminally located chlamydospores, some in short two to four membered chains. The hyphae had become more varied in form and thickness, with here and there terminal club-like swellings. These structures, which on further growth reached a length of up to 40 μ, consisted of two or more cells, and had rounded blunt ends, thus resembling the typical macroconidia of this species as seen in stationary culture. After 17 days these structures were more numerous, with very thick walls and dense granular contents. Some of them showed swelling of the walls of the individual compartments thus transforming themselves into chains of chlamydospores. Similar shorter structures, usually one- or two-celled, could be seen arising laterally from some hyphae. Arthrospores were fairly numerous and very irregular in shape. After 4 to 5 weeks there was marked lysis of the vegetative hyphae.

Discussion.

All the six dermatophyte species investigated seemed to develop in the same manner. Partly branching hyphae with and without segmentation were the first elements to appear. In some hyphae densely filled and empty segments alternated. Segmented hyphae were usually thicker than the non-segmented ones and their thickness further increased when arthrospore formation was initiated. All the species propagated mainly by two types of spores: arthrospores and chlamydospores. Arthrospore formation was the same in all species. After an initial increase in hyphal thickness regularly spaced moniliform swellings appeared and crosswalls were formed at the constrictions. The spores rounded off and finally chains of fairly round even spores were formed. Macroconidia-like structures were formed in all organisms. With *E. floccosum* and *T. tonsurans* they appeared early and were very numerous. With *T. mentagrophytes* and *T. rubrum* they were notably few in number and appeared at a late stage of the growth. Polymorphism was very marked with *T. mentagrophytes* and even more so with *T. schoenleini*. This latter organism possessed some structures not seen in any of the others, practically identical with the structures seen in the saprophytic phase of this fungus. The large number of chlamydospores and their wide range of shape and size were the most dominant features in the cultures of *T. violaceum*.

Differences in rate of growth were regularly found. The most rapid growth and earliest maturity occurred with *T. mentagrophytes*, followed in descending order by *T. tonsurans*, *T. rubrum*, *T. schoenleini*, *T. violaceum* and *E. floccosum*. The last three organisms grew at almost the same speed. Early lytic changes however were most marked with *E. floccosum.*

The difference in morphology between the "pseudoparasitic" and the saprophytic phase was most marked with *T. mentagrophytes*, *T. rubrum* and *T. tonsurans*, and much less so with *T. violaceum*, *E. floccosum* and *T. schoenleini*. With the latter organism the structures appearing in the two phases were identical. The most outstanding feature however was the total absence of microconidia in the pseudoparasitic phase of all organisms possessing them in their saprophytic phase, and of spirals in the growth of *T. mentagrophytes* and *E. floccosum.* Thus in its early growth the pseudoparasitic phase of all species resembled the morphology of dermatophytes as seen in clinical lesions of tinea. Further incubation with continuous shaking, however, produced in all cases more complicated structures not found in the parasitic phase. These later appearing structures were similar or identical with those found in the saprophytic life phase in stationary culture.

SUMMARY

The morphology of the pseudoparasitic life phase (continuous shake culture) of several dermatophytes is described. There was a strong resemblance between the forms developing in the early stages of this phase and those of the parasitic phase as found in clinical lesions. Propagation of the organisms mainly took place by the breaking up of hyphae into numerous arthrospores. During the later stages of development, however, structures not found in the parasitic phase appeared but which resembled those found in the saprophytic phase (stationary culture). This was especially true for *E. floccosum*, *T. violaceum* and *T. schoenleini*. The complete absence of microconidia in all species, and of spirals in *T. mentagrophytes* and *E. floccosum* was noted.

In addition to the different rates of growth certain microscopic features, most pronounced with *T. violaceum*, *T. schoenleini* and *E. floccosum*, seemed to permit differentiation of the various species in their pseudoparasitic life phase.

Reference

RAUBITSCHEK, F. (1955) Nutritional Requirements of Dermatophytes in Continuous Shake Culture. J. Invest. Dermat. **25:** 83—87.

ZUR MORPHOLOGIE DER PILZELEMENTE IM STRATUM CORNEUM BEI TINEA (EPIDERMOPHYTIA) PEDIS, MANUS ET INGUINALIS

von

Hans Götz

(*Aus der Dermatologischen Klinik und Poliklinik der Universität München* (*Direktor*: *Prof. Dr.* A. Marchionini)

(Mit 16 Textabbildungen)

(2.X.1959)

Im Gegensatz zu den bedingt möglichen Folgerungen auf Grund gewisser, aus pilzkranken Haaren zu erhebender Befunde, lassen sich bei einem Pilzbefall der Haut durch Dermatophyten Rückschlüsse auf die Art des ursächlichen Erregers nicht ziehen. Die Kalilaugenpräparate zeigen, daß die pathogenen Fadenpilze im Stratum corneum nur verhältnismäßig monomorphe Strukturen hervorbringen, nämlich mehr oder weniger zahlreiche Hyphen, die im wechselnden Maße zu Arthrosporen zerfallen. Nach Einführung der PAS-Färbung war die Möglichkeit gegeben, durch einen nicht sehr zeitraubenden Arbeitsgang zahlreiche Hautschnitte auf die Gegenwart von Pilzelementen zu untersuchen. Wie wir beobachteten, waren diese aber in ihrem Erscheinungsbild doch nicht so einförmig, wie wir zunächst annahmen.

TECHNIK

Zunächst bestimmten wir im Kalilaugenschuppenpräparat, ob ein im Sinne einer Tinea verdächtiger Krankheitsherd an der Fußsohle, im Handteller oder in der Leistenbeugenregion auch tatsächlich Pilzfäden enthielt. Bei positivem Ergebnis exzidierten wir im Flachschnitt unter Lokalanaesthesie ein etwa kleinfingernagelgroßes Epidermisfeld, das an seiner zentralen, also dicksten Stelle bis ins Corium hineinreichte. Das so erhaltene Biopsiematerial teilten wir durch Längsschnitte in 3 etwa gleich große Stückchen. Das erste verarbeiteten wir zur Kultur, um den ursächlichen Erreger zu züchten und zu klassifizieren. Das zweite und dritte Stück betteten wir sofort in üblicher Weise in Paraffin ein und stellten Serienschnitte von 10 bzw. 20 μ Dicke her:

a) im Vertikalschnitt
b) im Horizontalschnitt.

Abschließend erfolgte die Färbung dieser Schnitte mit der PAS-Methode zur Darstellung der Pilzelemente.

Auf diese Weise haben wir Epidermismaterial aus mykotischen Fuß-, Hand- und Inguinalläsionen untersucht und an Hand von über 1000 Einzelschnitten bestimmte Pilzstrukturen gefunden, die aber z. T. von der geltenden Vorstellung abweichen.

ERGEBNISSE

In allen Fällen handelte es sich um mehrjährige Pilzinfektionen, bei denen Besserungen mit Verschlechterung abwechselten. Klinisch lag an den Händen bzw. Füßen jeweils die squamös-hyperkeratotische Form vor. In allen Präparaten fanden wir Myzelfäden und typische Arthrosporen, doch auch Besonderheiten, die wir im Folgenden an Hand von 6 ausgewählten Fällen beschreiben. Die Abbildungen entsprechen einer 270-, 450- oder 950-fachen mikroskopischen Vergrößerung. Die wechselnde Bildvergrößerung wurde indessen nicht eigens vermerkt.

Fall I:

Exzisionsstelle: Großzehenballen
Gezüchteter Pilz: *Trichophyton rubrum*

Bei einer Durchmusterung aller Schnitte fallen in den äußersten Lagen des Stratum corneum lokalisierte, völlig abgerundete Sporen auf, die wegen ihrer Größe an einzeln liegende Chlamydosporen einer Kultur denken lassen und die wir als „makroide" Sporen bezeichnen wollen. Daneben finden sich aber auch kleine runde Gebilde, die den Mikrokonidien der Kultur entsprechen würden und von uns „mikroide" Sporen genannt werden. In der Abb. 1 ist ein solch makroides Element dargestellt, das in seinen Größenmaßen etwa den parasitären Haarsporen der makroiden Tricho-

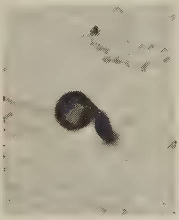


Abb. 1: 450fache Vergr. Makroide Spore (7,5 μ Durchmesser) des *Trichophyton rubrum* in den äusseren Zellagen des Stratum corneum des Großzehenballens.

phyten entspricht. Von der einen Durchmesser von 7,5 μ aufweisenden Spore geht ein kurzer, offenbar auskeimender Myzelschlauch ab, der an seinem freien Ende wiederum ein kleines kugeliges Gebilde aufweist (∼ 1,5—2,5 μ im Durchmesser). Hervorzuheben ist, daß wir in den vorliegenden Präparaten wiederholt auf solche winzigen Myzelkügelchen treffen, die meist am Ende eines kurzen Fadens lokalisiert sind (Abb. 2), aber auch isoliert gefunden werden (Abb. 3). Von einer 5 μ im Durchmesser betragenden Spore gehen drei Myzelschläuche ab, deren unteres Ende eine rundliche Anschwellung von 4 μ besitzt (Abb. 4). In deren Umgebung isoliert liegende, länglich-ovale Arthrosporen weisen eine Breite von 5—8 μ und eine Länge von 8—10 μ auf.

In den Abb. 5 und 6 ist ein interessanter Vorgang dargestellt. Offenbar kann es im Verlauf der Alterung bestimmter Pilzfäden auch zu einer Zusammenziehung des Protoplasmas an umschriebenen Stellen kommen. Die von der Anschwellung distal gelegenen Myzelteile sind rötlich-blaß, nur die Myzelhüllen scheinen noch

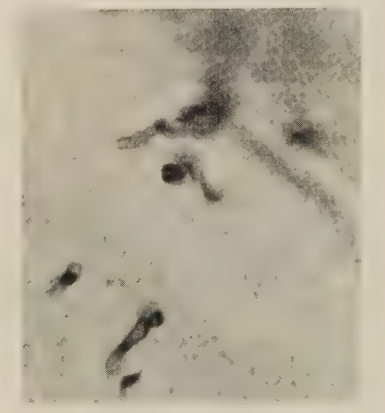


Abb. 2: 450fache Vergr.. Mikroide Spore (1,5—2,5 μ Durchmesser) des *Trichophyton rubrum* am Ende von Myzelschläuchen.

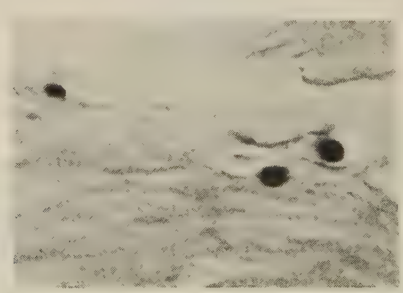


Abb. 3: 950fache Vergr.. Isoliert in den äußeren Zellagen des Stratum corneum liegende mikroide Sporen von der Größe der Abb. 2.

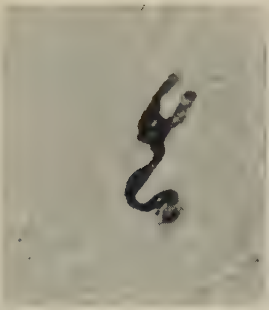


Abb. 4: 450fache Vergr.. 3 Myzelschläuche, von einer makroiden Spore des *Trichophyton rubrum* ausgehend.

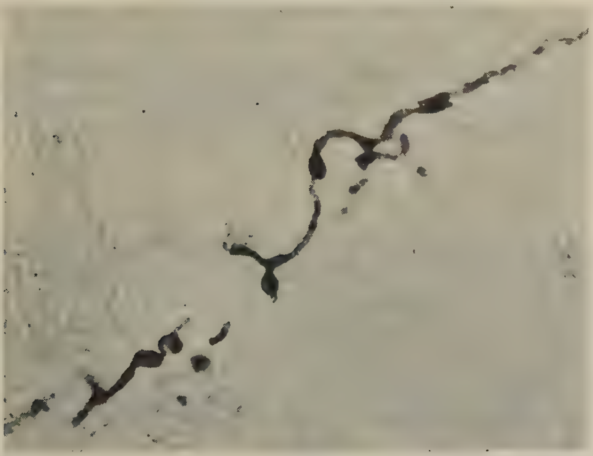


Abb. 5: 270fache Vergr.. Interkaläre Aufblähungen (5—6 μ breit) und Retraktion des Protoplasmas in einem Pilzfaden des *Trichophyton rubrum*.

vorhanden, die proximalen Anteile indessen zeigen in Übereinstimmung mit den großen kugeligen Gebilden eine tief blaurote Farbe. In der Abb. 5 sehen wir mehrere interkaläre Aufblähungen im Verlauf eines Pilzfadens, die jeweils 5—6 μ breit und 7,5—8 μ lang sind. Die stellenweise im Bereich der Septen einer Hyphe erfolgenden Myzelanschwellungen gemahnen an Bilder, die wir in der Kultur als Racquettemyzel bezeichnen. Die Dicke des normalen Filamentes schwankt zwischen 1,5—2,5 μ. In der Abb. 7 zeigt sich ein bizarres Myzelbild, das durchaus einem in vitro Kulturbefund entsprechen könnte. Wiederum erkennen wir umschriebene Myzelanschwellungen bis zu 5 μ Breite und 8—10 μ Länge neben mikroiden Sporen bis zu 2,5 μ Durchmesser. Bemerkenswert ist auch die Aufdeckung einer offenbar erfolgten

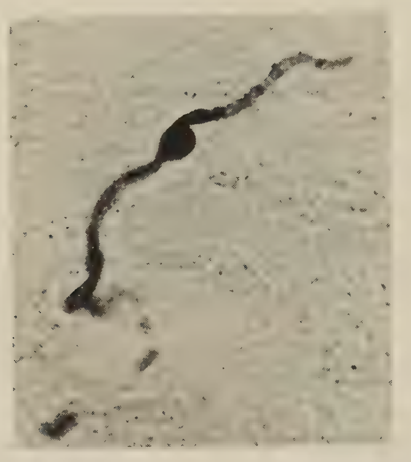


Abb. 6: 450fache Vergr.. Sporenbildung durch Retraktion des Protoplasmas (*Trichophyton rubrum*).

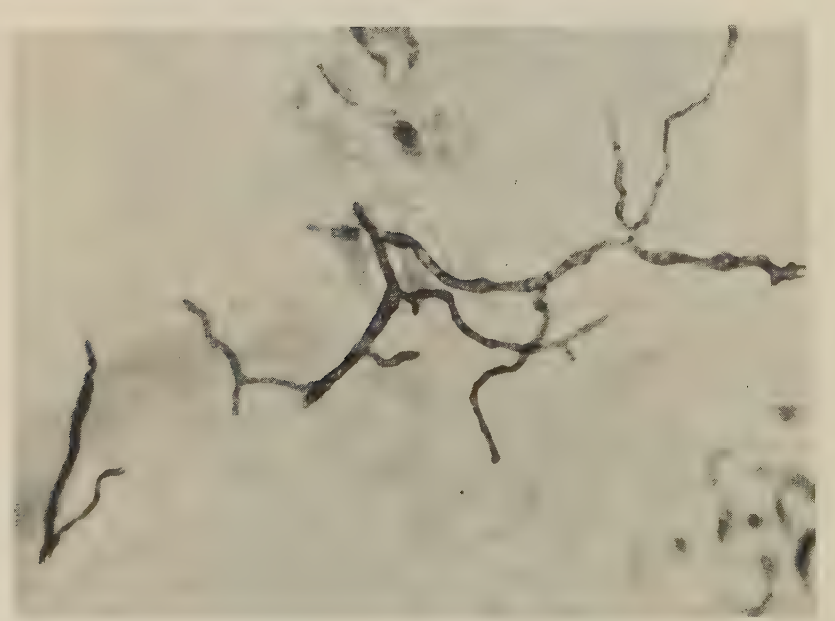


Abb. 7: 450fache Vergr.. Bizarres Myzelgeflecht des *Trichophyton rubrum* in den äußersten Zellagen des Stratum corneum, an Kulturbilder erinnernd.

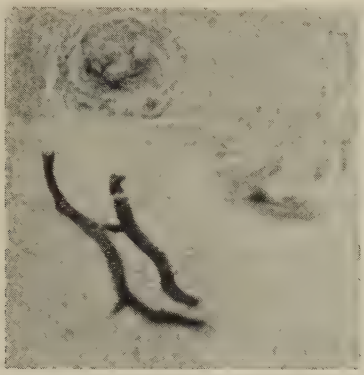


Abb. 8: 450fache Vergr.. Hyphenverschmelzung im Stratum corneum (*Trichophyton rubrum*).

Hyphenverschmelzung zwischen zwei 3 μ breiten Filamenten nach Art der Zapfen zu Zapfenanastomose, wie sie VAN UDEN im Objektträgerversuch beschrieben hat (Abb. 8). Das Verbindungsstück ist 1,5 μ breit.

Fall II:

Exzisionsstelle: Fußsohle
Gezüchteter Pilz: *Trichophyton rubrum*

In diesen Schnitten befinden sich Pilzfäden vielfach nur in der Tiefe, d. h. im unteren Drittel des 700 μ dicken Stratum corneum.

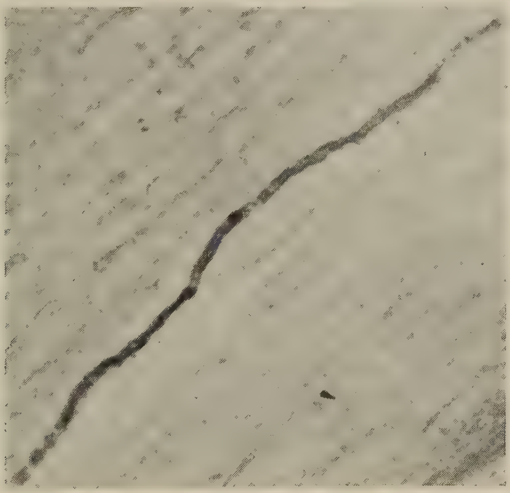


Abb. 9: 450fache Vergr.. Junger Pilzfaden des *Trichophyton rubrum* in der Tiefe des Stratum corneum der Fußsohle (blaß-rötlich gefärbtes Protoplasma, weiträumig entfernte dünne Septen).

Dabei zeigen diese ein nur blaß-rötlich gefärbtes Protoplasma, entwicklungsmäßig fast einen Kümmerwuchs. Ein umso intensiveres bläulich-rotes Kolorit tritt auf, je höher das Myzel zur Oberfläche zu liegen kommt. Da sich alte Pilzelemente stärker anfärben als junge, sind demnach die jüngsten Hyphen in der Tiefe der Hornschicht lokalisiert. Die im Verlauf der Hyphenreifung erfolgende Versporung in Richtung von der Tiefe zur Oberfläche bei horizontalem Wachstum der Fäden (diese werden passiv durch die kontinuierliche Regeneration der Hornschicht nach außen befördert), geht auch aus dem Verhalten der Septierung hervor (Horizontalschnitte). Der in der Abb. 9 wiedergegebene Faden in der Tiefe enthält nur 6 dünne, weiträumig entfernte Septen, das Protoplasma erscheint homogen, während ein älterer, oberflächlicher Faden deutlich dickere und engere, d. h. ältere Querwände aufweist (Abb. 10). Ein reifer, oberflächlich gelegener Myzelfaden wurde z. B. 185 μ lang befunden, mit 40 dicken Querwänden versehen.

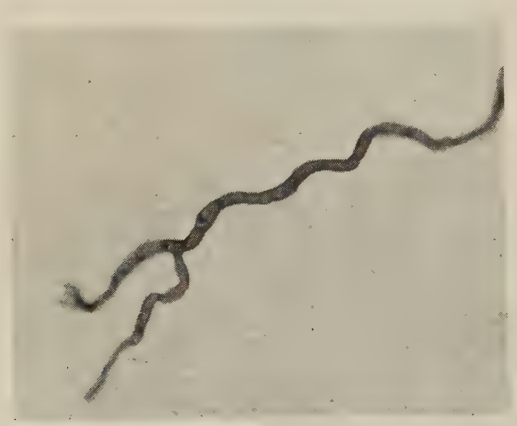


Abb. 10: 450fache Vergr.. Älterer Pilzfaden des *Trichophyton rubrum* im oberen Stratum corneum der Fußsohle (intensiv blau-rötliches Kolorit, dicke Querwände in kurzem Abstand).

Der Zerfall in eckige Arthrosporen, die an anderen Stellen durchschnittlich 4,5—5 μ Länge besaßen, war noch nicht eingetreten. Aber auch coriumwärts in die Tiefe führendes Hyphenwachstum läßt sich im Vertikalschnitt durch das Verhalten des blaßeren, noch nicht septierten, also jüngsten Myzelendes erkennen, während der in den höheren Hornschichten gelegene Myzelteil granulierter, dunkler erscheint, dicke Septen besitzt und somit älter ist.

Fall III:

Exzisionsstelle: Handfläche
Gezüchteter Pilz: *Trichophyton rubrum*

Das Stratum corneum ist an der Exzisionsstelle ca 600 μ dick. In den ersten Horizontalschnitten, die gewißermaßen nur das Stratum disjunctum der Hornschicht erfassen, sehen wir reichliche, isoliert im Keratin liegende Sporen, die teils rundlich, 5 μ groß, teils oval, 3 μ : 5 μ groß sind. Bisweilen haften einzelnen oder mehreren Hornplättchen winzige Myzelteilchen an, wie aus der Abb. 11 zu erkennen ist (isolierte längliche Arthrospore ca 11 μ lang neben Keratinplättchen). Aber auch völlig freie, ohne im Kontakt mit Keratinteilchen befindliche Pilzelemente können wir in typisch blau-roter Färbung auf dem Objektträger aufdecken. So erblicken wir wiederum winzige Sporen von 2,5 μ Länge und 1,5 μ Breite. Daneben auch große, ebenfalls isoliert liegende, fast runde Sporen (Maße 6 μ : 6,5 μ). In den äußersten Hornlagen ermitteln wir Sporenketten von bizarrer Konfiguration, deren Einzelglieder etwa 6 μ lang sind und z. T. an der Seite winzige Knospen, offenbar Stellen beginnender Auskeimung, tragen. Beispielsweise ist eine ausgereifte Hyphe rechteckig septiert, weist aber auffallend unterschiedliche Wandabstände auf (17,5 μ—10 μ—6 μ—15 μ—20 μ—5 μ—15 μ—5 μ—9 μ—5 μ—27,5 μ—12,5 μ). Bei der Durchmusterung der Vertikalschnitte imponieren im allgemeinen Myzelfäden mit rechteckigen Septierungen, bisweilen zeigen sie aber in kürzeren Abschnitten auch mehr Perlschnurcharakter. Die einzelnen, an den Ecken abgerundeten Sporen sind beispielsweise bei einem kurzen Faden im Durchmesser 5 μ, 4 μ, 4 μ, 4 μ, 3 μ und 5 μ groß.

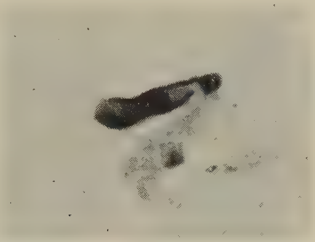


Abb. 11: 950fache Vergr.. 11 μ lange Arthrospore des *Trichophyton rubrum* im Stratum disjunctum der Handfläche, an einem Keratinplättchen haftend.

Fall IV:

Exzisionsstelle: Handfläche (exzidiert nach 14tägiger Pinselung mit Solutio Castellani, 2 × täglich)
Gezüchteter Pilz: *Trichophyton rubrum*

Aus den Schnitten geht hervor, daß unter dem Einfluß der antimykotischen Tinktur die oberflächlichen Pilzelemente geschädigt werden. Diese Beeinträchtigung der Entwicklung äußert sich teils in Einrissen der Pilzwände, die wie ausgefranst erscheinen, teils in geblähten oder geschrumpften kurzen Hyphenabschnitten. Manche Pilzfäden zeigen ein spitz zulaufendes Ende. Ähnliche mikroskopische Bilder konnten in pilzhaltigen mazerierten Hornteilchen des IV. Zehenzwischenraumes gefunden werden, aus denen die Pilzzüchtung aber nicht mehr gelang. Bisweilen stoßen wir auf Befunde, die an vertrocknende Hyphen erinnern. Die in früheren Schnitten des gleichen Patienten vor der Behandlung gefundenen, isoliert liegenden makroiden Sporen sind in den oberflächlichen Hornschichten fast nicht mehr nachweisbar. Nur in der Tiefe stoßen wir gelegentlich noch auf die im Vorhergehenden schon beschriebenen mikroiden Sporen von etwa 2 μ Durchmesser. Immerhin erscheint der Nachweis von geschädigten Myzelteilen selbst in der Tiefe der Hornschicht (aber neben noch immer gesunden, guten Turgor aufweisenden Hyphen!) bemerkenswert.

Fall V:

Exzisionsstelle: Inguinalregion
Gezüchteter Pilz: *Trichophyton rubrum*

Diese Präparate zeichnen sich auf Grund der veränderten Lokalisation durch ein nur durchschnittlich 30 μ dickes Stratum corneum aus. Im Gegensatz zu den bisherigen Myzelbildern besitzen die Filamente, obwohl es sich um die gleiche Pilzart handelt, eine sofort in die Augen springende beachtliche Dicke von durchschnittlich 3,5 μ (bis zu 4,5 μ). Sie wachsen vielfach parallel zur Oberfläche, seltener auch senkrecht in die Tiefe in Form kurzer Hyphenschläuche. Der Septenabstand schwankt in den Fäden zwischen 8 μ und 3,6 μ. Hier liegen unterschiedliche Reifegrade der Pilzhyphen vor, denn je älter die Filamente sind, umso mehr Querwände pflegen sich zu bilden und umso kleiner werden die Abstände. So zeigt z. B. ein junger, erst etwa 2,5 μ dicker Pilzfaden weite Septenabstände von 13 μ bis zu 45 μ bei sonst homogenem Inhalt. In auffallender Weise beinhalten in der Inguinalregion die gebildeten reifen Arthrosporen jeweils bis zu 3 stark lichtbrechende

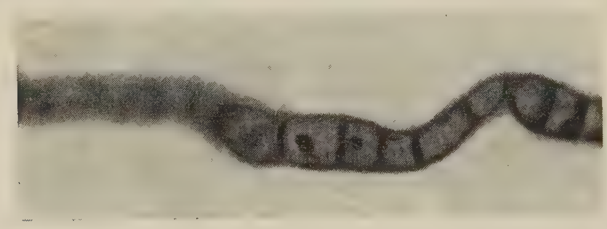


Abb. 12: 950fache Vergr.. Kurzer Pilzfaden des *Trichophyton rubrum* aus der Inguinalregion, dessen Arthrosporen merkwürdige Körperchen von polymorpher Begrenzung beinhalten.

Körperchen, die bei 950-facher mikroskopischer Vergrößerung rundlich, oval bis polymorph erscheinen. In der Abb. 12 ist ein solch charakteristischer Befund wiedergegeben. Die Arthrosporen sind eckig, zwischen 3—5 μ lang. Die einzelnen Körperchen liegen entweder isoliert im Zentrum oder längs der Wand, bzw. in einer Ecke, oder sie konfluieren. Ob es sich um Kristallisationsprodukte handelt, ist noch unklar. Auch wissen wir nicht, warum wir sie nur in der Hornschicht der Inguinalregion fanden.

Bizarr gewachsenes Myzel finden wir hier in gleicher Weise wie in manchen früheren *Trichophyton rubrum*-Präparaten. Wiederum begegnen wir den schon erwähnten kolbigen Protoplasmaanschwellungen, die durch Retraktion des Zellinhaltes zustande kommen, bis zu einem Durchmesser von 5 μ. Manche Hyphen dringen mit Sicherheit auch über das Stratum corneum hinaus in die Tiefe bis in das Stratum granulosum, z. T. sogar in das Stratum spinosum vor. Solche Befunde konnten wir nur dann erheben, wenn die Hornschicht verhältnismäßig dünn ist, also auch am Handrücken in einem anderen Fall, von dem die Abb. 13 stammt. Das *Trichophyton rubrum* war an mehreren Stellen bis in das Stratum spinosum vorgewachsen. Niemals aber fanden wir in den zahlreichen Schnitten Pilzelemente im Corium. Durch Verlagerung von locker liegenden Myzelteilchen bei der Herstellung der Serienschnitte kann es allerdings zu Trugschlüssen kommen, wie wir selbst beobachteten. So entdeckten wir einen stark septierten, mit einer Seitensprosse versehenen Myzelrest 440 μ tief im Corium in völlig reaktionsloser Umgebung. Die fast quadratischen, durchschnittlich 3,5 × 5 μ großen Arthrosporen enthielten je ein eingeschlossenes Körperchen der gleichen Art, die wir in der Hornschicht gefunden hatten und im Vorhergehenden beschrieben. Es handelt sich offensichtlich

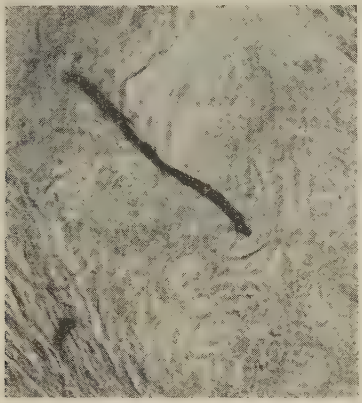


Abb. 13: 270fache Vergr.. Bis in die obersten Zellagen des Stratum spinosum (Handrücken) vordringender Pilzfaden des *Trichophyton rubrum*.

um artifiziell durch das Mikrotommesser verlagertes, reifes Myzel vom Typus der Abb. 11.

Andererseits liegt aber kein Kunstprodukt vor, wenn wir die Abb. 14 betrachten. Hier sehen wir Pilzfäden, die tief in einem mit Hornlamellen angefüllten Follikel der Inguinalregion vorgedrungen sind. In den Nachbarschnitten der Serie läßt sich die Gegenwart der Pilzelemente laufend verfolgen.

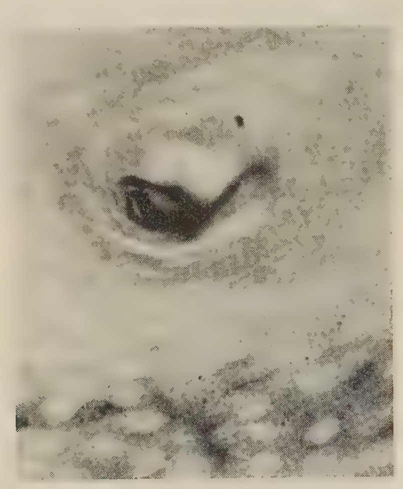


Abb. 14: 450fache Vergr.. Pilzgeflecht des *Trichophyton rubrum* tief in Follikelhornlamellen der Inguinalregion.

Fall VI:

Exzisionsstelle: Fußsohle

Gezüchteter Pilz: *Trichophyton mentagrophytes.*

Die Hornschichtdicke beträgt 760 μ. Eine Überprüfung aller Schnitte läßt reichliche, relativ gering verzweigte Myzelfäden erkennen (Abb. 15), die in den oberflächlichen Keratinlagen nahezu alle septiert sind. In der Abb. 16 ist eine dichotomisch verzweigte Hyphe wiedergegeben, deren Arthrosporen durchschnittlich 7,4 μ Länge aufweisen. An anderen Stellen berechnet sich nach Auszählung zahlreicher Arthrosporen ihre Durchschnittslänge auf 5,4 μ. Bizarren Myzelgebilden begegnen wir nicht. Gelegentlich zeigt sich ein Faden, dessen noch zusammenhängende Arthrosporen bereits wieder kurze 3—4 μ lange Keimschläuche nach verschiedenen Seiten vortreiben. Eine mikroide Rundspore von 2,5 μ Durchmesser kann einer solchen beginnenden Seitensprosse aufsitzen. Die Hyphen in der Tiefe des Stratum corneum sind in gleicher Weise wie bei einem *Trichophyton rubrum*-Befund blaß-rosa. gefärbt.

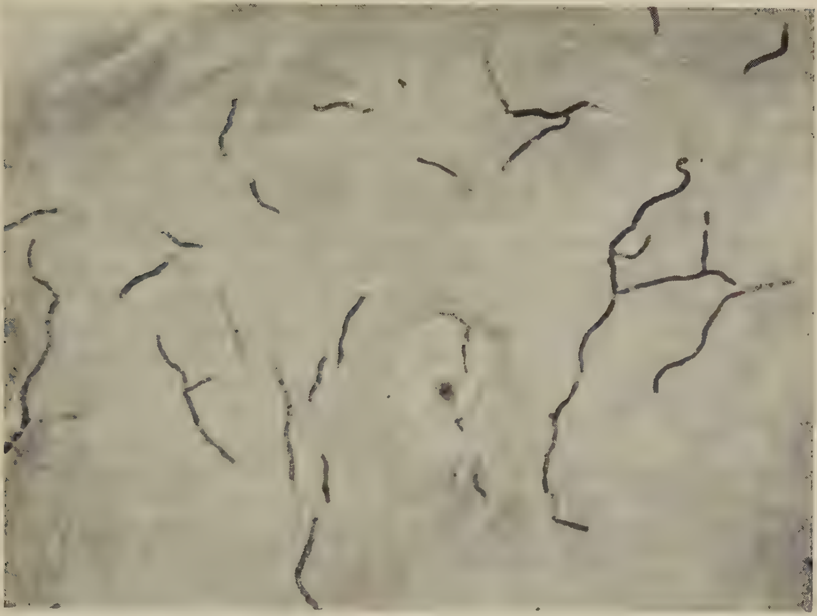


Abb. 15: 270fache Vergr.. Zahlreiche, wenig verzweigte, aber sehr stark septierte Pilzfäden des *Trichophyton mentagrophytes* im Stratum corneum der Fußsohle.

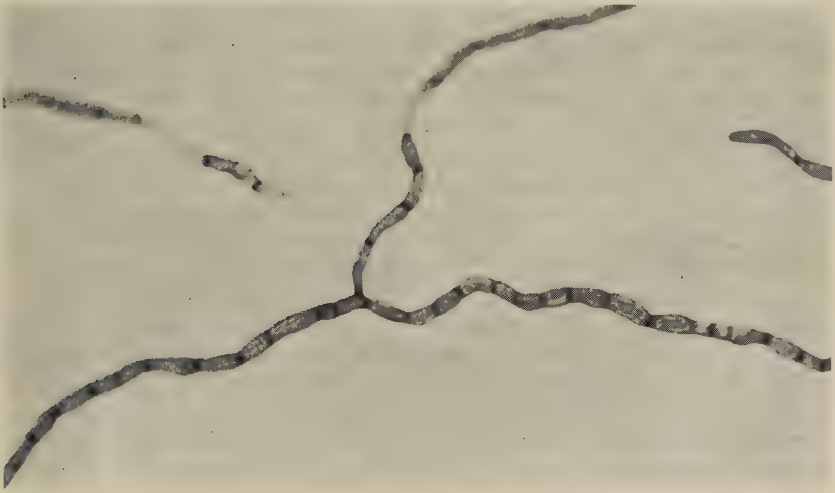


Abb. 16: 450fache Vergr.. Reifer *Trichophyton mentagrophytes*-Pilzfaden; Arthrosporen durchschnittlich 7,4 μ Länge.

BESPRECHUNG DER ERGEBNISSE

Trichophyton rubrum

Dieser Pilz wurde aus dem Material der als Beispiele dienenden Fälle I und II (Fußsohlenhaut), III und IV (Handflächenhaut) und V (Epidermis der Inguinalregion) gezüchtet. Wenn sich auch, wohl durch Lokalisationsfaktoren bedingt, gewisse Abweichungen der Befunde ergaben — (die Stärke der Hyphen betrug in dem durchschnittlich 600 μ dicken Keratinder Hand- und Fußsohlen nur 1,5 μ—2,5 μ, in dem dünnen Keratin der Inguinalregion hingegen vorwiegend 3 μ—4,5 μ. Offenbar herrschen hier bessere Ernährungsbedingungen, denn das inguinale Myzel darf man geradezu als „fett" bezeichnen. Vielleicht spielt die bessere O_2-Versorgung eine Rolle, ist doch das Stratum corneum hier nur etwa 30 μ dick. Auch sahen wir nur in der Leistenbeugengegend stark lichtbrechende, in den Arthrosporen eingeschlossene kernartige Körperchen) —, so ließen sich andererseits doch Gemeinsamkeiten finden. In den oberflächlichen Hornlagen der Fußsohlen und Handflächen wiesen wir mehr oder weniger zahlreiche makroide rundliche Sporen von 5 μ bis zu 7,5 μ Größe nach, neben mehr als „mikroid" zu bezeichnenden runden Gebilden, die meist kurzen Myzelsprossen aufsaßen (1,5—2,5 μ Größe). In der Hand-, Fuß- und Inguinalregion zeigte das Protoplasma der Pilzfäden die Tendenz, sich an umschriebenen Stellen zusammenzuziehen und ~ 5 μ dicke kolbige Anschwellungen nach Art der Chlamydosporenbildung zu entwickeln. Vereinzelt entstanden Bilder, die mit Racquettemyzelien vergleichbar sind. Da diese Anschwellungen etwa die gleiche Größe wie die isoliert in den Hornlagen zu findenden makroiden Sporen besitzen, scheint uns zumindest ein Teil dieser Elemente durch Protoplasmaverschiebungen gebildet zu werden. Bizarr konfigurierte Myzelgeflechte erinnerten bisweilen an saprophytische Kulturbilder. Ein zweiter Modus der Sporenbildung erfolgt in der bekannten Weise durch Zerfall des Myzelfadens in rechteckige, bisweilen auch an den Ecken abgerundete Arthrosporen mit nicht selten stark wechselnden Septenabständen. In den tiefen Lagen des Stratum corneum der Handflächen und Fußsohlen findet sich vorwiegend junges Myzel (schwache Färbung, keine oder unterentwickelte Septenbildung), das, während es durch die kontinuierliche Regeneration der Hornschicht oberflächenwärts abgeschoben wird, allmählich heranreift. An umschriebenen Stellen ließ sich der Pilz nur im unteren Drittel der Hornschicht nachweisen, während die oberen pilzfrei erschienen. Das erklärt bisweilen die negativen Pilzbefunde bei oberflächlicher Schuppenabnahme, obwohl klinisch kein Zweifel an dem Vorliegen einer Pilzflechte zu bestehen scheint.

Trichophyton mentagrophytes

Es wurde aus dem als Beispiel dienenden Fall VI (Fußsohlenhaut) gezüchtet. Die Tendenz zur Arthrosporenbildung war allgemein stärker ausgeprägt. Wenn sich auch hinsichtlich der Maße (5 μ—7μ) keine klaren Abgrenzungen gegenüber den mehr rechteckigen Arthrosporen des *Trichophyton rubrum* aufdecken ließen, so fiel doch in allen Schnitten auf, daß im Keratin zwar viele Hyphen schmarotzten, diese aber in den oberflächlichen Lagen in ihrer überwiegenden Mehrzahl septiert sind. Bizarr konfigurierte Myzelgebilde entdeckten wir nicht. Ebenso vermißten wir die bei dem *Trichophyton rubrum* beschriebenen kolbigen Anschwellungen im Verlauf einiger Pilzfäden. Schon Kalilaugenpräparate mit Schuppen von *Trichophyton mentagrophytes* (*granulosum*)-Fällen ließen fast immer jene verstärkte Tendenz zur Versporung der Myzelfäden erkennen.

In keinem Fall ermittelten wir ein Myzelgeflecht, das aus in allen Dimensionen sich kreuzenden, miteinander verschlingenden Hyphen bestand, wie wir es aus Kulturbildern kennen. Zwar deckten wir bisweilen umschriebene Stellen auf, die zahlreiche Pilzfäden enthielten (vor allem bei dem *Trichophyton mentagrophytes*), doch stellten wir an Hand unserer Horizontal- und Vertikalschnitte fest, daß die Pilzfäden untereinander einen regeren Kontakt nicht zu suchen schienen. Man hatte vielmehr den Eindruck, jedes einzelne Filament verfolge sein eigenes Ziel, d. h. kommt seiner eigenen „Futtersuche" nach. Das schließt aber offenbar gelegentliche Strukturen im Sinne einer Hyphenverschmelzung (Abb. 7) nicht aus.

Allgemeines

Die im Falle IV (Handflächenhaut nach 14-tägiger Pinselung mit Solutio Castellani) erhaltenen Schnitte ließen eine Schädigung der Pilzfäden in Form platzender Wandungen, einer Aufsplitterung der Myzelenden und daher spitz auslaufender Fäden erkennen. Auch konnten wir die oft kurzen Myzelenden aufsitzenden mikroiden Sporen fast nur noch in den tiefen Keratinschichten ausmachen. Bemerkenswert ist unseres Erachtens, daß wir solcherart geschädigte Pilzelemente — wenn allerdings auch nur an einigen Stellen — selbst in den tiefen Schichten des Stratum corneum fanden. Entweder handelt es sich hier um spontane Schädigungen (die wir manchmal auch in Präparaten unbehandelter Kranken entdecken konnten), oder das in der Solutio Castellani enthaltene Phenol dringt wahrscheinlich bis in die tiefen Hornlagen ein. Möglicherweise lassen sich daher bei monatelanger konsequenter Pinselung mit dieser antimykotischen Tinktur auf die Dauer doch alle Pilzelemente erreichen und vernichten. Leider scheitert dieses Ziel im allgemeinen an der bald nachlassenden Behandlungsausdauer der Patienten.

In der Inguinalregion (auch am Handrücken in einem Fall) kam es zu gelegentlichen Befunden, die bis in das Stratum spinosum vordringende Myzelfäden zeigten. Pilzbefunde im Corium konnten aber als Artefakte erkannt werden. Eine Erklärung für die nicht seltene Therapieresistenz der Tinea inguinalis ließ sich insofern finden, als wir Pilzfäden tief in einem keratingefüllten Follikel nachwiesen. Die meisten Antimykotika dringen hier, extern appliziert, nur schwer in die Tiefe, so daß von einer solchen Infektionsquelle ausgehend jeweils wieder unter günstigen Bedingungen ein Rezidiv aufzuflammen vermag.

Auch im Hinblick auf prophylaktische Maßnahmen erhielten wir einige Einblicke. Wenn wir bedenken, in welch großer Zahl in den oberflächlichen Horizontalschnitten des Biopsiematerials I (Fußsohlenhaut) und III (Handflächenhaut) locker verstreut liegende Arthrosporen sowie die beschriebenen mikroiden und makroiden Sporen (teils an einzelnen Hornplättchen haftend) nachgewiesen wurden, vermögen wir eine Vorstellung zu gewinnen von der ständigen Gefahr, die solche Kranken für ihre Mitmenschen darstellen. Ein einfacher Händedruck genügt, zahlreiche winzige Pilzsporen auf die Handfläche des Partners zu übertragen. In gleicher Weise verliert ein fußpilzkranker Patient ständig zahlreiche Sporen in seine Umgebung. Einige von uns erhobene weitere Befunde sprechen dafür, daß erfreulicherweise diese parasitären Pilzsporen weniger vital sind als saprophytische Kultursporen. So gehen sie im trockenen Milieu sehr viel rascher zugrunde als Kultursporen. Wir werden an anderer Stelle auf diese Beobachtungen zurückkommen.

ZUSAMMENFASSUNG

Durch flache, bis ins Corium reichende Exzisionen wurde Epidermismaterial aus mykotischen Fuß-, Hand- und Inguinalläsionen mit Hilfe der PAS-Färbung auf morphologische Besonderheiten der ursächlichen Dermatophyten überprüft.

1. Die Hypen des *Trichophyton rubrum* entwickelten im durchschnittlich 600 μ dicken Stratum corneum der Hand- und Fußsohle eine Stärke von 1,5 μ—2,5 μ, in der nur etwa 30 μ betragenden Hornschicht der Inguinalregion eine Stärke von 3 μ—4,5 μ.
2. Bei einem Teil der *Trichophyton rubrum*-Arthrosporen aus der Inguinalregion fanden wir im Zellinneren vorwiegend 1, manchmal aber bis zu 3 stark lichtbrechende, teils rundliche, teils polymorphe Körperchen, deren Deutung noch unklar ist (Kristallisationsprodukte?).
3. Wurde aus dem Stratum corneum ein *Trichophyton rubrum* gezüchtet, so ließen sich in den oberflächlichen Hornlagen zwei Arten von Sporenbildungen beobachten.
   a) durch Zerfall eines Pilzfadens in mehr rechteckige, aber auch in rundlich-ovale Arthrosporen,
   b) durch Anschwellung umschriebener Hyphenstellen infolge Retraktion des benachbarten Protoplasmas.

   Die Neigung des *Trichophyton rubrum* zur Bildung bizarrer

Myzelstrukturen verdient hervorgehoben zu werden. Eine Hyphenverschmelzung ließ sich einmal finden.

4. Das *Trichophyton mentagrophytes* hatte in der Hornschicht zahlreiche Pilzfäden mit geringer Verzweigungstendenz gebildet, die aber in den oberflächlichen Keratinlagen nahezu alle nach dem Modus der Arthrosporenbildung septiert waren. Diese Neigung trat weit stärker hervor als bei dem *Trichophyton rubrum.*
5. Sowohl bei dem *Trichophyton rubrum* als auch dem *Trichophyton mentagrophytes* stießen wir wiederholt auf 1,5—2,5 μ kleine runde Kügelchen, die meist kurzen Myzelenden aufsaßen. Ob diesen Gebilden die Eigenschaften von Sporen zu kommen, ist noch offen.
6. Die jüngsten Hyphen parasitierten in der Tiefe der Hornschicht. Sie waren teils noch unseptiert bzw. wiesen nur sehr dünne Querwände und ein blaß-rosafarbenes Kolorit auf. Sie fanden sich bei dem Trichophyton rubrum abschnittsweise nur im unteren Drittel des Stratum corneum. Die älteren, reifen Pilzfäden waren kräftig gefärbt und durch dicke Querwände gut septiert.
7. Vom Stratum disjunctum aus werden Pilzsporen teils an Keratinplättchen haftend, teils auch frei in die Umgebung verstreut. Die Vitalität dieser parasitären Sporen ist geringer als die der saprophytischen Kultursporen.
8. Gegenwart von Myzelfragmenten im Corium konnte mechanisch durch Verschleppung vom Stratum corneum aus bei der Herstellung der Serienschnitte erklärt werden. Mit Sicherheit lassen sich aber an Körperstellen mit dünner Hornschicht (Handrücken, Leistenbeuge) Pilzfäden nachweisen, die bis in die obersten Zellagen des Stratum spinosum vordringen. An Handinnenflächen und Fußsohlen konnten wir solche Befunde nicht erheben.
9. Ein Faktor für die Chronizität inguinaler Tinea-Infektionen ließ sich in aufgefundenen Hyphengeflechten tief im Follikelhals finden.

Summary

By serial sections extending into the corium we obtained epidermal material from mycotic lesions of the foot, hand and inguinal regions. These we stained by the PAS-reaction for morphological pecularities of the causative dermatophytes.

1. In the stratum corneum of the palms and soles which had a thickness of about 600μ, the hyphae of the *Trichophyton rubrum* developed a diameter from 1.5μ—2.5μ. In the stratum corneum of the inguinal region which was only 30μ thick, we found a diameter of 3μ—4.5μ.

2. In some of the arthrospores of the *Trichophyton rubrum* in the inguinal region we noticed in the cellular substance highly re-

fractile, partly round, partly polymorphous bodies ranging from predominantly 1, sometimes to 3 in number. The meaning of these is not yet clear (products of cristallization?)

3. When we cultured *Trichophyton rubrum* from the stratum corneum, we observed two modes of spore formation in the superficial horny layers:

a) By division of a fungous filament in mostly rectangular, but also in roundish and oval arthrospores.

b) By swelling of localized sites of hyphae due to the retraction of adjacent protoplasma.

The tendency of the *Trichophyton rubrum* for the formation of bizarre structures of the mycelium deserves to be mentioned. Once a hyphal fusion was noted.

4. The *Trichophyton mentagrophytes* in the horny layer had developed numerous filaments with little tendency for branching. In the superficial layers however almost all of these filaments were septate according to the mode of arthrospore formation (division). This tendency was much more marked with *Trichophyton mentagrophytes* than with *Trichophyton rubrum.*

5. In both *Trichophyton rubrum* and *Trichophyton mentagrophytes* we noted repeatedly small round balls of 1.5μ—2.5μ diameter, almost always situated on top of short hyphae. Whether these structures are genuine spores is not yet clear.

6. The youngest hyphae grew in the deepest part of the horny layer. Some of them were still non-septate or they showed thin septums and a faint rose-coloured tint. In cases of *Trichophyton rubrum* hyphae were found in localized areas to be situated only in the lower third of the stratum corneum. The older and more mature filaments were deeply coloured and well divided by thick septums.

7. Fungus spores will be scattered from the stratum disjunctum, some of them adhering to horny particles, some of them free of all skin attachments. These parasitic spores are more sensitive to environmental influences than spores derived from a culture.

8. The presence of fungus fragments in the corium could be explained by mechanical introduction from the stratum corneum during serial sectioning of the specimen. In areas with thin horny layers (dorsum of the hands, inguinal region) we could demonstrate with certainty hyphae extending to the uppermost layers of the stratum spinosum. We could not implicate such findings on the palms and soles.

9. One factor explaining the chronicity of the tinea-infection in the inguinal region is the finding of fungus filaments deep in the follicle.

Literatur

UDEN, N. VAN, Arch. Derm. Syph. (Berlin) **193,** 468 (1951/52).

TWO NEW SPECIES OF PLEOSPORA FROM INDIA

S. T. TILAK

(*M. A. C. S. Laboratory, Law College Buildings, Poona* 4, *India*).

(19.X.1959)

During the course of his mycological investigations on the Ascomycetes of the Bombay State, the writer collected two fungi, one from dried wood and another from fresh leaves. The fungi were characterised by typical muriform golden brown ascospores and the presence of paraphysoids within the ascocarps. A critical study was, therefore, carried out in microtome sections, the results of which showed that the two fungi belonged to the genus, *Pleospora* as originally defined by RABENHORST (1857).

The fungus genus *Pleospora* according to RABENHORST (1857) is characterised by muriform brown to yellow coloured spores with paraphysoids, continuous from floor to bottom. SUTHERLAND (1916) does not mention paraphysis or paraphysoids in his description of *Pleospora pelvetiae*, although he describes the paraphyses as being deliquescent at maturity in *Pleospora laminaria*.

This genus is represented in the Indian Flora by only 3 species, while there is no record of this genus from Bombay State. SACCARDO (1883, 1902) describes 143 species of this genus. The genus has been recently revised by WEHMEYER (1948—1954) and MULLER (1951). The two collections made by the writer showed significant morphological differences in respect of the position of the ascocarp to the host tissue, the ascocarp was completely embedded in leaves of *Hiptage bengalensis* KURZ. while on the dried stems of *Cocculus villosus* DC. it was entirely superficial. These have been, therefore, presented below as new to science on the basis of host relationship and morphological characters.

(1) *Pleospora mullerii* TILAK sp. nov.

Ascocarps dark, completely embedded, uni-locular, globose, 119—170 = 136—221 μ, breaking at maturity with a round pore. Stroma consists of layers of cells, thick walled and inner layers somewhat thin walled. Asci in basal layers, numerous, cylindrical to clavate, bitunicate, 8-spored, 68—90 = 17—19 μ. Paraphysoides filiform. Ascospores somewhat biseriate, muriform with 3 transverse septa and 2—3 vertical septa, golden brown, 6—7 celled, thick walled, 17—21 = 6—9 μ. Collected in the months of July-August 1958. Law College Hills, Poona on living leaves of *Hiptage bengalensis* KURZ. Leg. S. T. TILAK.

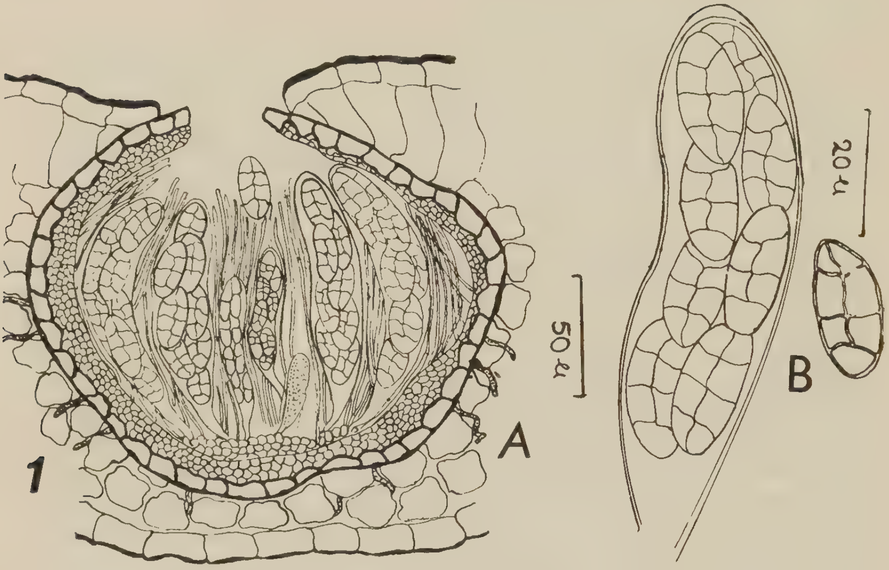


Fig. 1. *Pleospora mullerii*
A. Section through ascocarp. B. Ascus and ascospores.

Ascocarpi fusci, penitus immersi, uniloculares, globosi, 119—170 = 136—221 μ, ad maturitatem dehiscentes poro circulari. Stroma contans seriebus cellularum crassis perietibus praeditarum, et seriebus internis cellularum nonnihil tenuibus parietibus praeditarum. Asci basaliter seriati, plurimi, cylindrici vel clavati, bitunicati, octospori, 68—90 = 17—19 μ. Paraphysoides filiformes. Ascosporae aliquantum biseriatae, muriformes, trino transverso, bino vel trino verticali septo distinctae, aurae bruneae, 6—7-cellulatae, crassis parietibus praeditae, 17—21 = 6—9 μ. Typus lectus mensibus julio et augusto anni 1958 ad Law College College Hills, in urbe Poona, in foliis viventibus *Hiptages bengalensis* KURZ. Leg S. T. TILAK.

The species has been described after Dr. EMIL MULLER of the Department of Special Botany, Swiss Federal Institute of Technology, Zurich in recognition of his many contributions to the Ascomycetous fungi.

(2) *Pleospora arxii* TILAK sp. nov.

Ascocarps dark, entirely superficial, uni-loculate, flattened, ovoid, ostiolate, 85—100 = 153—187 μ. Stroma consist of 2—3 layers of cells outermost thickened. Asci in basal layer, numerous, clavate, pedicillate, bitunicate, 8-spored, 60—70 = 15—19 μ. Paraphysoides continuous enveloping the asci. Ascospores biseriate when young but somewhat irregular at maturity, with 3 cross and two vertical septa, 6-celled, golden brown to dark brown, 18—20 = 6—8 μ, collected on the stem of *Cocculus villosus* DC.

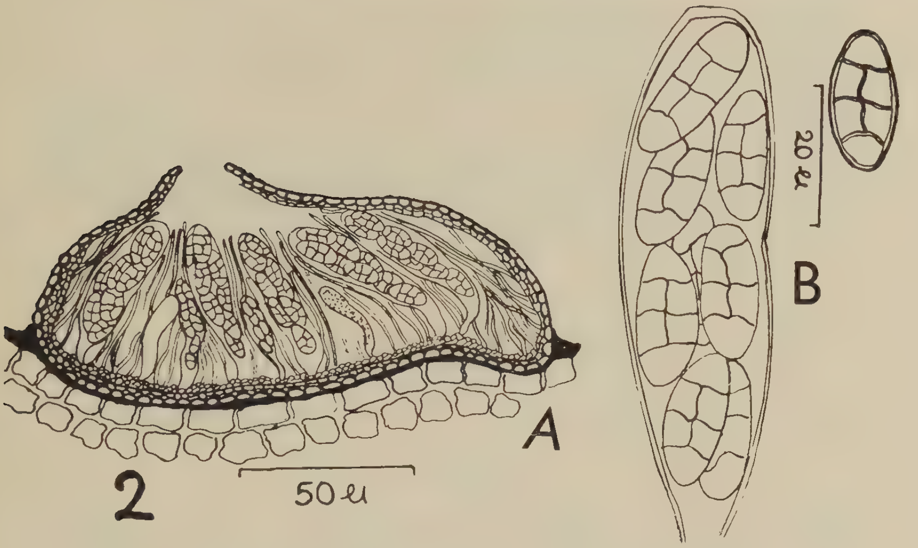


Fig. 2. *Pleospora arxii*
A. Section through ascocarp. B. Ascus and ascospores.

20th June, 1959, Law College Hills, Poona. Leg. S. T. TILAK.

Ascocarpi fusci, penitus superficiales, uniloculati, complanati, ovoidei, ostiolati, 85—100 = 153—187 μ. Stroma constans duplici vel triplici serie cellularum, quarum externae crassae. Asci basaliter seriati, plurimi, clavati, pedicellati, bitunicati, octospori, 60—70 = 15—19 μ. Paraphysoides continuae ascum involventes. Ascosporae biseriatae in statu juvenili, aliquantum irregulares ad maturitatem, septis 2 transversis, 2 vero verticalibus ornatae, 6-cellulatae, auree bruneae vel vusce bruneae, 18—20 = 6—8 μ, lectae in culmis *Cocculi villosi* DC. die 20 junii anni 1959, ad Law College Hills, in urbe Poona. Leg. S. T. TILAK.

The species has been described after Dr. J. A. VON ARX of the Phytopathological Institute, Baarn, Netherlands in recognition of his many contributions to the Ascomycetous fungi.

Besides being new species, the genus is a new addition to the Bombay Flora.

The type specimens have been deposited at the Herbaria of the Commonwealth Mycological Institute, Kew, England and Indian Agricultural Research Institute, New Delhi, India.

The writer is grateful to Prof. M. N. KAMAT for guidance and encouragement, to Dr. S. P. AGHARKAR for facilities offered at the M.A.C.S. Laboratory. He is also thankful to Rev. Father H. SANTAPAU for Latin diagnosis.

References

MULLER, EMIL, (1951) Die Schweizerischen Arten der Gattungen Clathospora Pleospora, Pseudoplea und Pyrenophora. Sydowia **5**: 248—310.

RABENHORST, L. (1857) Herbarium Myc. eidt. II, 347.

SACCARDO, P. A. (1883) Sylloge Fungorum Vol. II, p. 241—277.

SACCARDO, P. A. (1902) Sylloge Fungorum Vol. XVI, p. 544—548.

SUTHERLAND, G. K. (1916) Additional notes on Marine Pyrenomycetes. Trans. Brit. Myc. Soc. **5**: 260.

WEHMEYER, L. E. (1948) The development pattern in the genus *Pleospora* Rab. Mycol. **40**: 269—294.

WEHMEYER, L. E. (1949) Studies in the genus *Pleospora* I. Mycologia **41**: 565—593.

WEHMEYER, L. E. (1951) Studies in the genus *Pleospora* III. Mycologia **44**: 34—53.

WEHMEYER, L. E. (1953) Studies in the genus *Pleospora* V. Mycologia **46**: 391—414.

NUTRITIONAL STUDIES OF EIDAMELLA DEFLEXA. I. CARBON-NITROGEN ASSIMILATION AND SOME GROWTH FACTOR REQUIREMENTS.

by

Harold H. Kuehn & Paul F. Crosby

Grain Processing Corporation, Muscatine, Iowa, U.S.A.

(1.IX.1959)

The developmental morphology of *Eidamella deflexa* (Berk.) Benjamin was reported upon by both DeLamater (1937) and Kuehn (1956). In addition, Benjamin (1956) and Kuehn (1956, 1958) reviewed the synonomy of this species and listed the following as synonyms of *E. deflexa*: *Myxotrichum deflexum* Berk., *Eidamella spinosa* Matruchot and Dassonville, and *Eidamella papyricola* Saccas. Dodge (1935) considered *Gymnoascus setosus* Eidam to be a synonym of *Eidamella deflexa* but DeLamater (1937) presented a comparison of the two species and concluded that they were distinct. However, DeLamater was not able to induce his culture of *G. setosus* to produce ascocarps and his conclusions must therefore be regarded as unconfirmed.

In a previous report, Kuehn & Crosby (1959) reviewed the literature regarding physiological work with representatives of the Gymnoascaceae. There have been several such publications regarding *Gymnoascus setosus*, notably those of Hawker (1936), Schopfer & Blumer (1940) and Nyuksha (1953, 1957). Their work will be reviewed briefly because of the possibility that *G. setosus* is actually conspecific with *Eidamella deflexa*. Hawker cultured *G. setosus* on an agar medium containing 0.5 % glucose, 0.35 % KNO_3, 0.075 % $MgSO_4$ and 0.175 % KH_2PO_4 to which addition of either a crude lentil extract or an inositol-free fraction of the crude extract failed to improve the fruiting of the fungus. Schopfer & Blumer reported that *G. setosus* grew on a mineral-dextrose medium which contained asparagine. The addition of thiamin or its intermediates had no effect on the growth of the fungus. Nyuksha studied *G. setosus* in more detail and reported that pigmentation and growth occurred normally on sugars (only feebly on lactose), starch and cellulose but not on citric acid. The best source of nitrogen was peptone while in descending order the mold also used gelatin, KNO_3 asparagine and $(NH_4)_2SO_4$. He also reported that the fungus grew well on a medium which lacked a nitrogen source, although on such a medium no pigment was elaborated. NYUKSHA performed several experiments with regard to factors affecting the production of a red pigment by his fungus. According to DELAMATER (1937) the mycelium of *G. setosus* is colorless and he claimed that this characteristic could be used to distinguish *G. setosus* from *E. deflexa*, at least on certain media. Furthermore, NYUKSHA isolated his strain from paper, a common habitat for *E. deflexa*. Therefore, it seems likely that NYUKSHA's fungus in reality was *E. deflexa*.

No previous information has been published concerning the nutrition or physiology of *Eidamella deflexa* with the possible exception of the reports of NYUKSHA. However, GHOSH (1954) studied the physiology of certain Gymnoascaceae, including *E. deflexa*. She found that his fungus utilized dextrose, fructose, galactose, maltose, sucrose, raffinose, dextrin, lactose and xylose as carbon sources but not sodium lactate. She also found that asparagine was the best nitrogen source, and that glycine, casein hydrolysate, $NaNO_3$ and an amino acid mixture also supported growth, while no growth resulted with the use of NH_4NO_3 or $(NH_4)_2SO_4$. Furthermore, she found that zinc and copper were essential for growth. Comparison of the results of NYUKSHA and GHOSH shows certain similarities which would indicate that they were working with the same species.

MATERIALS AND METHODS

The fungus used in this study was *Eidamella deflexa* (BERK.) BENJAMIN RSABG 57 (Rancho Santa Ana Botanic Garden culture number). This strain had been isolated by R. K. BENJAMIN from an old board in a basement, Belmont, Massachusetts, USA, in March, 1952. Since 1953 this fungus has been maintained by one of the authors (HHK) on YpSs and Sabouraud's agar media.

The constituents of YpSs (0.4 % yeast extract, 0.1 % K_2HPO_4, 1.5 % soluble starch, 0.05 % $MgSO_4{\cdot}7H_2O$) and Sabouraud's (4 % glucose or maltose, 1 % peptone) media were used in preliminary experiments. Various carbon and nitrogen-containing compounds were substituted in these media in order to determine the best carbon and nitrogen sources for growth. The concentrations employed varied from one experiment to another, and therefore the levels of the constituents of the media will be reported in conjunction with the individual experiments. The experimental procedure, methods of determining growth, etc., were similar to those explained in detail previously (KUEHN & CROSBY, 1959) and the reader is referred to that article for further information. In general, the following procedure was employed. Five hundred ml Erlenmeyer flasks containing 100 ml medium were inoculated with 2 % of 4—10 day old submerged growth brought up in Sabouraud's maltose broth. The flasks were incubated on a reciprocating shaker making 93 complete 4½ inch strokes per minute. Preliminary experiments showed that the fungus failed to grow at 37° C, so all experiments were conducted at 28° C. The amount of growth was determined by centrifuging the contents of the flasks, washing the mycelium twice with water, and drying the mycelium to a constant weight overnight in a constant-temperature oven at 100° C. All weights reported represent the average of at least two replicate flasks.

A red pigment is readily produced by this fungus on agar slants of YpSs and other media. This red pigment also was produced in the shaken flasks. Intensity of pigmentation was determined photometrically after it had been demonstrated that there was a linear relation between the amount of the culture filtrate and per cent transmission as recorded on a Bausch and Lomb Spectronic 20 colorimeter at a wave length of 450 mu.

RESULTS AND DISCUSSION

Preliminary experiments were conducted to determine the effect of the pH range from 2.8—10.0 on growth of the fungus. Using Sabouraud's glucose medium with a 13 day incubation period optimum growth was obtained with an initial pH of 7.0. This experiment was repeated over a narrower pH range using a 6 day incubation period and the results are shown in Table I. It was evident that the initial pH within the range of 6—8 was not critical for growth or pigment elaboration.

Several replicate flasks containing the YpSs medium at pH 7.0 were placed on the shaker simultaneously to determine the rate

TABLE I.

Effect of initial pH upon the growth of Eidamella deflexa grown on Sabouraud's glucose medium

Flask	Initial pH	Final pH	Mycelial weight mg/100 ml	% Transmission 1 : 10*
1	6.0	4.5	555	36
2	,,	4.5		43
3	6.5	4.5	576	32
4	,,	4.5		31
5	7.0	4.6	577	28
6	,,	4.5		31
7	7.5	4.6	559	29
8	,,	4.4		38
9	8.0	4.5	545	37
10	,,	4.4		25

* Indicates dilution of culture filtrate. Readings were made using 16 × 150 mm test tubes.

of growth and the optimum incubation time. At daily intervals two flasks were removed from the shaker and the amount of growth was determined. The results indicated that maximum growth was attained between 4—6 days and thereafter the weight of mycelium decreased with the onset of autolysis. Table II presents the results of a second experiment conducted to verify the initial results.

TABLE II.

Rate of growth of Eidamella deflexa in YpSs medium.

Days of incubation	Final pH	Mycelial weight mg/100 ml
1	6.9	159
2	5.9	255
3	5.5	477
4	4.8	515
5	4.4	517
6	4.4	558
7	4.8	460
8	5.0	398
9	5.2	333

TABLE III.

Growth of Eidamella deflexa on different sources of carbon in a peptone medium.

Compound	Final pH	% Transmission 1 : 10	Mycelial weight mg/100 ml
lactose	5.1	17	1022
soluble starch	4.2	17	779
maltose	4.2	9	755
sorbitol	5.3	9	746
glucose	4.2	14	557
sucrose	4.5	36	535
dextrin	4.4	23	505
mannose	4.5	39	456
fructose	4.7	54	409
inositol	8.5	87	271
calcium gluconate	8.5	91	178
sodium acetate	8.2	89	161
adipic acid	8.2	86	158
propionic acid	7.4	93	141
oxalic acid	7.1	89	107
raffinose	8.6	83	87
malic acid	8.5	92	79
sorbose	5.1	—	75
sodium citrate	8.1	90	64
itaconic acid	8.5	92	60
xylose	5.3	—	46
citric acid	7.6	91	41
succinic acid	8.5	80	40
lactic acid	4.9	92	26
control	8.7	91	15

Peptone was used as the nitrogen source in the following carbon utilization studies. The levels of ingredients were those which were used in the stock Sabouraud's agar slants. The pH of each flask was adjusted to 7.0 following addition of the carbon compound. After an incubation period of 7 days, the results shown in Table III were obtained. The compounds are listed in descending order of availability. Since lactose permitted considerably more growth than any other carbon source, several subsequent experiments were conducted using lactose as the source of carbon. However, consistent pigment elaboration never was achieved with lactose as a carbon source. Although cellular yields were consistently high, pigment intensity varied from almost none to an intense red. Table IV shows the results of an experiment in which the experimental design was a duplicate of that described for Table III. In each experiment lactose supported more growth than the other test compounds; however, in one experiment lactose permitted a high degree of pigmentation while in the other almost no red pigment was present. No cause or explanation could be found for this behavior of lactose when used as the carbon source with peptone as the nitrogen source. Further studies are planned to elucidate the effect of lactose. It is interesting to note that NYUKSHA (1957) reported only slight growth for his fungus using lactose as the carbon source.

TABLE IV.

Additional data on carbon utilization by Eidamella deflexa

Compound	Final pH	% Transmission 1 : 10	Mycelial weight mg/100 ml
lactose	6.7	80	874
maltose	4.7	12	796
sucrose	4.2	20	764
sorbitol	6.0	4	700
glucose	4.4	20	637
soluble starch	4.4	44	531
dextrin	8.6	75	203
control	8.6	88	77

Eidamella deflexa had been maintained on agar slants of YpSs, Sabouraud's and Czapek's media, which contained yeast extract, peptone and $NaNO_3$ respectively as the nitrogen sources. In order to study the nitrogen assimilation of this fungus various nitrogen-containing compounds were substituted at a concentration of 0.4 % for yeast extract in the YpSs medium. The flasks were adjusted to pH 7.0 following addition of the nitrogen compound and incubated for 6 days. Table V presents the results with the compounds listed in descending order of utilization. It can be seen that KNO_3 was the best source of nitrogen among those compounds which were tested.

TABLE V.

Nitrogen assimilation of Eidamella deflexa with soluble starch as the carbon source.

Compound	Final pH	% Transmission 1 : 10	Mycelial weight mg/100 ml
KNO_3	4.6	60	443
NH_4 citrate	5.8	69	389
$Ca(NO_3)_2$	5.9	52	365
$NH_4H_2PO_4$	5.2	54	364
$NaNO_3$	6.0	67	341
urea nitrate	6.4	54	330
NH_4 tartrate	5.5	52	291
yeast extract	3.8	40	249
NH_4 lactate	5.5	52	240
asparagine	5.5	60	239
NH_4Cl	4.4	75	223
dl-aspartic acid	5.6	65	222
urea oxalate	7.7	54	212
peptone	3.9	50	188
NH_4NO_3	4.9	71	187
sodium asparaginate	4.5	14	185
corn steep liquor	4.3	38	176
choline citrate	5.5	69	171
dl-phenyl alanine	3.9	54	164
urea	7.3	67	150
controls	3.9	79	150
arginine	5.1	70	126
methionine	4.0	57	122
beta alanine	5.4	77	121
isoleucine	3.4	56	120
NH_4HCO_3	6.9	73	120
choline chloride	3.6	76	119
glycine	3.6	42	115
valine	3.5	54	110
casein hydrolysate	3.8	70	110
controls *			
soluble starch	4.6	86	36
CMC	6.0	89	31

* These compounds, at a level of 1.5 %, were supplied singly and represented the only constituent of the medium.

In a further experiment regarding nitrogen assimilation, lactose was used as the carbon source at a 4 % level, while certain nitrogen-containing compounds were added at a 1 % level. After a 6 day incubation period the results (Table VI) showed that glycine was better than any other defined or complex nitrogen source. However, in attempting to reproduce these results considerable variation was encountered. Table VII shows some representative data of an attempt to verify the findings presented in Table VI. Again, no explanation could be found for the variation resulting from the use of lactose as the source of carbon.

Comparison of our results with those of NYUKSHA (1957) and GHOSH (1954) shows agreement in the non-utilization of $(NH_4)_2SO_4$.

TABLE VI.

Nitrogen assimilation of Eidamella deflexa with lactose as the carbon source.

Compound	Final pH	% Transmission 1 : 10	Mycelial weight mg/100 ml
glycine	5.3	21	581
yeast extract	5.2	30	554
corn steep liquor	4.8	48	543
casein hydrolysate	5.1	62	536
peptone	5.1	50	459
tryptophan	5.1	62	394
dl-histidine HCl	5.2	64	371
cystine	5.3	84	353
glutamic acid	4.6	32	288
KNO_3	4.4	78	237
lysine	5.0	63	186
$(NH_4)_2SO_4$	3.4	84	168
NH_4M4O_4	5.8	62	129
NH_4 acetate	5.9	85	124
urea	6.5	70	119

TABLE VII.

Further studies of nitrogen assimilation using lactose as the carbon source.

Compound	Final pH	% Transmission 1 : 10	Mycelial weight mg/100 ml
yeast extract	6.2	1	981
peptone	5.4	18	862
corn steep liquor	4.8	15	679
glycine	4.7	33	454
sodium asparaginate	4.8	29	322
KNO_3	4.7	79	297
NH_4 lactate	6.0	52	265

However, NYUKSHA reported good growth in the absence of a nitrogen source, a statement which we were unable to confirm since very little growth resulted in the presence of soluble starch or carboxymethylcellulose (CMC) alone.

Glycine consistently proved to be the best defined, single source of nitrogen with lactose as the carbon source, while KNO_3 was the best nitrogen source when soluble starch was the carbon source. Certain defined nitrogen compounds (glycine, KNO_3, dl-asparagine, NH_4 lactate) were tested in all possible combinations. A total of 1 % of the combined nitrogen sources was used. No combination was superior to glycine or KNO_3 alone.

In order to determine the effect of the carbon source upon nitrogen utilization, the best carbon sources at a 4 % level were tested with either 1 % glycine or 1 % KNO_3 for 7 days. The results shown in Table VIII illustrate that neither lactose nor soluble

TABLE VIII.

Growth of Eidamella deflexa as affected by combinations of carbon sources with KNO_3 and glycine.

Nitrogen source	Carbon source	Final pH	% Transmission 1 : 10	Mycelial weight mg/100 ml
KNO_3	maltose	5.7	11	539
	soluble starch	5.8	28	389
	sorbitol	5.2	84	303
	lactose	5.0	55	266
	glucose	5.0	46	264
	sucrose	5.2	50	228
glycine	sorbitol	7.0	42	583
	maltose	6.0	0	529
	soluble starch	6.9	51	435
	lactose	6.2	32	395
	sucrose	5.5	30	313
	glucose	5.7	40	251

starch represent the best source of carbon. Maltose was far superior to any other carbohydrate when KNO_3 was used and maltose was second only to sorbitol when glycine was the nitrogen source. However, maltose supported the maximum pigmentation with each nitrogen source.

Biotin, thiamin, calcium pantothenate and pyridoxine at levels of 0.2 mg % were added singly to test media of either maltose and glycine or maltose and KNO_3 to examine the effect of these vitamins upon growth. In addition 0.2 mg % biotin plus 0.2 mg % thiamin were supplied in additional test flasks of each medium. The highest yields of both cellular material and pigment were obtained in that medium which contained maltose-glycine plus the combination of 0.2 mg % each of thiamin and biotin. This stimulatory effect of thiamin is in direct contrast to the report of SCHOPFER & BLUMER (1940) regarding "*G. setosus*".

A further experiment to define the growth factor requirements of *E. deflexa* was performed by adding various purines and pyrimidines singly to a glycine-maltose test medium. The test compounds, adenine, xanthine, guanine and uracil, were added in concentrations of 1—8 mg %. After a 5 day incubation period the results showed that uracil at any of the test concentrations provided a stimulus for growth over the control yields. However, no stimulation of pigment production resulted with any of the test compounds or concentrations.

Two experiments were performed in attempts to verify certain findings which NYUKSHA (1957) reported for "*G. setosus*". He found that increased pigment elaboration resulted in the presence of these metals and concentrations: $CuSO_4 \cdot 5H_2O$, 0.0001 (% by

TABLE IX.

The effect of certain trace metals upon growth and pigmentation of E. deflexa.

Nitrogen source	Metal	Final pH	% Transmission 1 : 10	Mycelial weight mg/100 ml
glycine	Zn	4.4	14	196
	Cu	5.8	29	343
	Mo	5.8	51	141
	Fe	5.0	67	80
	Co	5.9	74	259
	control	4.7	33	454
KNO_3	Cu	4.9	65	275
	Fe	4.8	67	211
	Mo	5.2	70	132
	Zn	6.0	84	165
	Co	5.8	91	71
	control	4.7	79	297
NH_4lactate	Mo	6.0	66	194
	Cu	6.1	71	198
	Fe	5.5	71	88
	Zn	5.6	78	396
	Co	5.6	85	154
	control	6.0	52	265
peptone	Zn	5.6	6	876
	Mo	5.9	17	407
	Cu	6.4	28	720
	Fe	8.6	60	86
	Co	8.7	67	259
	control	5.4	18	862

weight metal concentration in the medium); $ZnSO_4 \cdot 7H_2O$, 0.005; $(NH_4)_2MoO_4$, 0.01, 0.02; $FeSO_4 \cdot 4H_2O$, 0.01; $CoCO_3$, 0.005. These same compounds and concentrations were used with the exception that $CoCl_2 \cdot 6H_2O$ was substituted for $CoCO_3$. The experiment was divided into four sets of flasks with the nitrogen source representing the only variant. The test medium consisted of 4 % lactose plus 1 % of the nitrogen source as represented by one of the following: glycine, KNO_3, NH_4 lactate or peptone. Following a ten day incubation period the results, shown in Table IX, revealed that only Cu and Fe in the lactose-KNO_3 medium and Zn in the lactose-peptone and lactose-glycine media significantly enhanced pigmentation. Nyuksha reported an increase in pigmentation of 4—6 fold with Mo, 3—3.5 fold with Co and 2—3 fold with Fe. Aside from the possibility that different fungi were being tested these differences could be attributed to different test media.

NYUKSHA used 1 % sucrose and 0.1 % peptone, which probably was a deficient medium. Also, our work was done in shaken flasks while NYUKSHA does not present his procedures in sufficient detail to permit definite conclusions to be drawn concerning his methods.

NYUKSHA further reported that darkness was more favorable for pigment elaboration. To examine his findings a test medium was used which consisted of 1 % glycine, 4 % maltose, 0.2 mg % biotin, 0.2 mg % thiamin and 8 mg % uracil with a pH of 7.0. After a 5 day incubation period there were no significant differences among flasks grown in bright light, subdued light or darkness. Table X presents the average values of a number of replicate flasks incubated under varying light conditions.

TABLE X.

The effect of light upon pigmentation and growth of E. deflexa.

Light condition	Final pH	% Transmission 1 : 100	Mycelial weight mg/100 ml
Darkness	4.3	67	692
Bright light	4.3	63	664
Subdued light	4.3	68	720

SUMMARY

Growth of *Eidamella deflexa* (BERK.) BENJAMIN was determined using increase in dry weight as an index of growth. The fungus was cultured in shaken flasks on a reciprocating shaker with 100 ml medium per 500 ml Erlenmeyer flask. Mycelium production was determined with 25 different carbon compounds and 37 different nitrogen sources which were supplied singly. It was found that the nature of the nitrogen source affected the response of the fungus to a given carbon source. Also, the nature of the carbon source affected response toward a given nitrogen source. Lactose, soluble starch, maltose and sorbitol were the carbohydrates which permitted maximum growth when peptone was the nitrogen source. Glycine was the best defined nitrogen source when lactose was the carbon source, while KNO_3 was the best defined nitrogen source with soluble starch as the carbon source. When glycine was the nitrogen source sorbitol and maltose were the best carbon sources, while maltose was the best carbon source with KNO_3 as the nitrogen source. Mixtures of defined nitrogen sources did not permit greater mycelium production than either glycine or KNO_3 alone.

The best initial pH ranged from 6.0—8.0 with the optimum around 6.5—7.0. The maximum production of mycelium resulted in 5 days at 28° C. The fungus was not able to grow at 37° C. Addition of 0.2 mg % biotin, 0.2 mg % thiamin and 8 mg % uracil increased mycelium production in a maltose-glycine or maltose-KNO_3 medium.

The effect of the various ingredients of the medium upon pigmentation was noted. Maltose elicited the best pigmentation among the carbohydrates, while the best nitrogen sources were yeast

extract, peptone and corn steep liquor followed by glycine and sodium asparaginate.

Trace metals such as zinc, copper and iron influenced pigmentation but the extent of this influence was affected by the nature of the nitrogen source. The amount of light did not affect pigment elaboration.

Literature Cited

Benjamin, R. K. (1956) A new genus of the Gymnoascaceae with a review of the other genera. Aliso **3:** 301—328.

DeLamater, E. D. (1937) *Eidamella spinosa* (Matruchot and Dassonville) refound. Mycologia **29:** 572—582.

Dodge, C. W. (1935) Medical Mycology. Mosby, St. Louis. 900 pp.

Ghosh, G. R. (1954) Comparative physiology of some members of the Gymnoascaceae. Doctoral Dissertation, University of Illinois, 64 pp.

Hawker, L. E. (1936) The effect of certain accessory growth substances on the sporulation of *Melanospora destruens* and of some other fungi. Ann. Bot. **50:** 699—719.

Kuehn, H. H. (1956) Observations on Gymnoascaceae. III. Developmental morphology of *Gymnoascus reessii*, a new species of *Gymnoascus* and *Eidamella deflexa*. Mycologia **48:** 805—820.

—, (1958) A preliminary survey of the Gymnoascaceae. I. Mycologia **50:** 417—439.

— & Crosby, P. F. (1959) Growth and nutrition of *Pseudoarachniotus roseus* in submerged culture. Mycopath. et Mycol. Appl. **11:** 109—127.

Nyuksha, Y. P. (1953) A physiological study of the fungus *Gymnoascus setosus* Eidam. I. Mikrobiologiya **22:** 15—22.

—, (1957) A physiological study of the fungus *Gymnoascus setosus* Eidam. II. Mikrobiologiya **26:** 309—315.

Schopfer, W. H. & Blumer, S. (1940) Recherches sur la répartition de l'hétérotrophie par rapport à l'aneurine chez les champignons. Arch. Microbiol. **11:** 205—214.

A NEW SPECIES OF MYCOSPHAERELLA FROM INDIA

by

T. S. Viswanathan

M. A. C. S. Laboratory, Poona-4, India

(25.VIII.1959)

In the course of his mycological collections, the writer encountered some plants of *Morus alba* L. in the vicinity of Poona, showing typical leaf spots. These infection spots on microscopical examination revealed the presence of a species of *Cercospora*. On a more thorough examination, the fungus was identified as *Cercospora moricola* Cooke. Leaves with older infection spots revealed the presence of an ascigerous stage with the typical characters of *Mycosphaerella*. Although many species of *Mycosphaerella* have been reported from other countries associated with the *Cercospora* stage on this host, this is the first time that an ascigerous stage has been observed on this host in India. A more critical study of this fungus was, therefore, undertaken in microtome sections, the results of which are presented in this paper.

Four different species of *Mycosphaerella* have, so far, been reported on *Morus alba* L. and one on *Morus rubra* L. A comparison between the Indian species of *Mycosphaerella* and the other five species is presented in the following table:

Comparison of species of Mycosphaerella on morus species

Species	Location of perithecium	Size of perithecium	Size of Asci	Size of Ascospores	Authority
Sphaerella morifolia	Hypophyllous	—	50—55 X 10—15 μ	17—25 X 5.0—5.5 μ	Pass.
Sphaerella Mori-albae	Hypophyllous	—	—	—	Cooke
Mycosphaerella moricola	Hypophyllous	36—75 X 32—71 μ	28—43 X 8—10 μ	16—18 X 3.5 μ	Sawada
Mycosphaerella mori	Epiphyllous	60—80 μ	35—40 X 5.5—6.5 μ	12—14 X 3.5—4 μ	Wolf
Mycosphaerella arachnoidea	Amphigenous, mostly epiphyllous	80—100 μ	45—54 X 7—8.5 μ	14—17 X 3.5—4 μ	Wolf
Indian species	Epiphyllous	60—70 μ	24—30 X 6—8 μ	9—12 X 2 μ	—

It is evident from the foregoing tabular statement that the Indian species is significantly distinct from the other five species in possessing smaller perithecia, asci and ascospores. Hence the Indian species is presented as a new species of *Mycosphaerella* with Latin diagnosis.

Mycosphaerella indica VISWANATHAN spec. nov.

Infection spots circular to irregular, 1—8 mm in diameter, with grey to tan center and black margin. Perithecia epiphyllous, dark brown, globose, 60—70μ in diameter. Ostiolate, ostiole erumpent measuring 7—12μ. Asci fasciculate, elongate-cylindric, 8-spored, aparaphysate, 24—30 × 6—8μ. Ascospores 1—2 seriate, hyaline, asymmetric, 1-septate, 9—12 × 2μ.

On living leaves of *Morus alba* L. Poona (India). by T. S. VISWANATHAN, 15-August 1958.

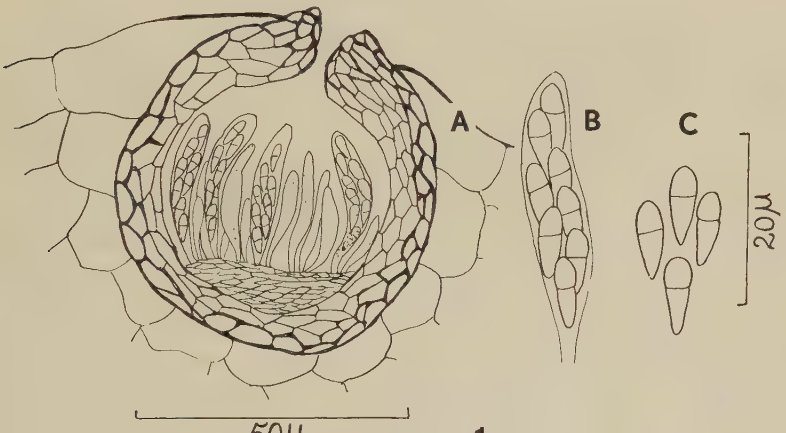


Fig. 1. *Mycosphaerella indica*:

A. Section through perithecium.
B. Ascus with ascospores.
C. Ascospores.

Mycosphaerella indica VISWANATHAN spec. nov.

Infectionis maculae circulares vel irregulares, 1—8 mm diam., centro griseo vel bruneo, marginibus nigris. Perithecia epiphylla, fusce brunea, globosa, 60—70μ diam., Ostiolata, ostiolo erumpente magnit. 7—12μ diam. Asci fasciculati, clavati vel cylindrici, ectospori, aparaphysati, 24—30 × 6—8μ. Ascosporae 1—2 seriatae, hyalinae, asymmetricae, 1-septatae, 9—12 × 2μ.

Typus lectus in foliis viventibus *Mori albae* L. ad Poona in India a T. S. VISWANATHAN die 15 augusti anni 1958.

The type specimens have been deposited in Herbarium Cryptogamae Indiae Orientalia, New Delhi, India and the Herbarium of the Commonwealth Mycological Institute, Kew, England.

The author wishes to express his grateful thanks to Prof. M. N. KAMAT for guidance, to Prof. S. P. AGHARKAR, Director, for providing laboratory facilities and to Rev. Father H. SANTAPAU for Latin diagnosis.

Reference

WOLF, F. A. (1936) False mildew of red mulberry. Mycologia, **28**: 266—277.

USE OF ACHROMYCIN IN THE PREPARATION OF SELECTIVE MEDIA FOR FUNGI

by

H. S. Andleigh,

M. D. (Path.), D. T. M., D. Bact., (Lond.)

Professor of Pathology and Bacteriology, S. M. S. Medical College, Jaipur, India.

The isolation of pathogenic fungi from clinical material, such as the sputum, skin scrapings, and the discharges from the sinuses etc., is not a simple and straight-forward laboratory procedure. The contaminating microorganisms grow so profusely that they do not allow the proper growth of pathogenic fungi.

Various techniques have been put forward from time to time with the object of suppressing the growth of saprophytic fungi and the contaminating bacteria. Several media have been described for the isolation of fungi from clinical material such as sputum which is usually heavily contaminated with bacteria. In 1942 Kurung used a dextrose-yeast extract medium of high acidity (pH 4.0) to inhibit bacterial growth. From this medium transfers were made to Sabouraud's agar for identification. In 1945, Thompson described the undesirable aspects of Sabourauds medium with and without crystal violet. He used successfully a hormone blood agar (pH 7.0) to which he added 2 units of penicillin and 10 units of streptomycin per ml of the medium. This supported the growth of several pathogenic fungi and proved useful in isolating *Coccidioides immitis* from sputum in pure culture. A selective medium for *Coccidioides immitis* (1 % ammonium chloride, 1 % Sodium acetate, 0.8 % tribasic potassium phosphate, 0.4 % cupric sulphate and 2 % agar) was described by Smith in 1943. In 1947 and 1948 Littman described an oxgall streptomycin crystal violet medium for the cultivation of fungi from contaminated clinical material. The medium not only restricted bacterial growth, but restricted the size of fungus colonies. This latter is of advantage in that the fast growing fungus contaminants i.e. *Aspergillus*, etc., do not overgrow most of the pathogenic fungi. *H. capsulatum* grow only after heavy inoculation. In 1948 Howell used a brain heart infusion blood agar with the addition of 20 units of penicillin and 40 units of streptomycin per ml of medium for the successful cultivation of *H. capsulatum.*

These days Brain Heart Infusion liquid medium or Brain Heart Agar with 20 units of penicillin and 40 units of stremptomycin per ml of the medium, is supposed to be the best medium for the isolation of most of the pathogenic fungi. In the beginning of my work in mycology in 1950, I started using media meant for growing pathogenic fungi. to which penicillin and streptomycin was added in the concentration of 20 units of penicillin and 40 units of streptomycin, respectively. This worked nicely for the isolation of some pathogenic fungi. Later on Actidione was discovered.

Actidione is an antibiotic which is very active against many yeasts and fungi but is tolerated in relatively high concentrations by most bacteria. Actidione may be added to bacteriological media to facilitate the isolation of pathogenic fungi or counting of bacteria in the presence of yeast or molds.

A medium containing 100 micrograms of Actidione per ml has been reported to be useful in the isolation of pathogenic fungi from clinical lesions. Actidione proved of great help in the isolation of dermatophytes. The addition of 500 micrograms of actidione, 20 units of penicillin and 40 units of streptomycin per ml of Sabouraud's dextrose agar, that had previously been sterilised and cooled to 45° C, has been reported to make possible the isolation of *T. mentagrophytes*, *T. rubrum* and *Epidermophyton floccosum.* However, several pathogenic bacteria can be cultured on nutrient dextrose agar medium containing upto 1000 micrograms of Actidione. per ml of the medium. A medium containing Actidione Polymixin, Bacitracin, Inculin and Crystal violet has been reported to be more effective in inhibiting the growth of fungal contaminants.

During my work on the investigations into the etiology of Maduromycosis in India, I found that it was very difficult to isolate the fungi in pure culture as a type of Gram's negative bacillus belonging to the Friedlander group always contaminated the growth and did not stop growing on either the liquid media or the solid media containing penicillin and streptomycin. I then tried to find out the sensitivity of these organisms against the different antibiotics and found that they were sensitive to achromycin even in the concentration of 100 micrograms per ml of the medium. They were found to be most sensitive to Achromycin and less sensitive to Chloromycetin and much less sensitive to streptomycin and Penicillin. Achromycin was then used in the medium along with another set of media containing Penicillin and Streptomycin to find out as to which was a more suitable antibiotic. All sorts of clinical material such as the sputum and the skin scrapings were tried. The medium used for these investigations was prepared in our laboratories as follows.

37 g of Difcos dehydrated brain heart infusion powder was added to 1000 ml of distilled water in a 2 litre flask. The material was

dissolved and it was sterilised at 15 lbs. pressure for 30 minutes. Beside the liquid medium, Brain Heart agar medium was also prepared. To one set of the medium cooled to 35° C, penicillin and streptomycin was added in the proportion of 20 units of penicillin and 40 units of streptomycin per ml of the medium and to another set of the medium achromycin in the proportion of 500 micrograms of pure achromycin powder per ml of the medium was added. Actidione in the proportion of 100 micrograms per ml of the medium was added to both types of media. Primary inoculation of the material from cases of mycetoma foot were put up on the two sets of each of the Brain Heart Infusion liquid medium, and brain heart infusion agar slopes containing penicillin with streptomycin and achromycin respectively. Similarly sputum cultures of sinus discharges and scarpings were put up.

It was found that while on the media containing penicillin and streptomycin some bacterial contaminants appeared, particularly the gram negative bacilli in cases of Mycetoma cultures, no such bacterial contaminants appeared on the media containing achromycin. Achromycin has another advantage in being much more stable than penicillin besides its being a broad spectrum antibiotic. Moreover, penicillin and streptomycin act in a neutral or slightly alkaline medium. The pH of Sabouraud's dextrose agar is fairly on the acid side. Achromycin on the other hand works best in a acid medium. Achromycin is therefore more suitable to be incorporated in the most widely used Sabouraud's dextrose agar medium than any other antibiotic. Achromycin is thus the antibiotic of choice to be incorporated in the media used for the isolation of pathogenic fungi from all sorts of clinical material such as sputum, skin scrapings and sulphur granules etc.

Summary

The isolation of pathogenic fungi from heavily contaminated material has always been difficult uptil now. Brain Heart Infusion medium containing 30 units of penicillin and 40 units of streptomycin has been found to be the best medium for the isolation of pathogenic fungi. Better results have been obtained by using Achromycin in place of penicillin and streptomycin.

Zusammenfassung

Die Isolierung von pathogenen Pilzen aus schwer kontaminiertem Material waz bis jetzt schwierig. — Hirn-Herz — Infusion Medium, das 30 I.E. Penicillin and 40 I.E. Streptomycin enthält, ist als das beste Medium für die Isolierung pathogener Fungi gefunden worden. Bessere Resultate sind erzielt worden wenn Achromycin anstatt von Penicillin und Streptomycin verwendet wurde.

Acknowledgements

My thanks are due to Messrs Lederle Laboratory (India) Private Ltd., for supplying me Achromycin.

References

1. ANDLEIGH, H. S. (1951) Laboratory Diagnosis of Mycotic Lung Infections. J. Ind. med. Assoc., **20**: 360—371.
2. ANDLEIGH, H. S. (1957) In vitro study of antifungal activity of Pentamidine and Stilbamidine. Mycopathol. Mycol. Appl. **8**, 135—137.
3. ANDLEIGH, H. S. (1957) Etiology of Maduromycosis in India. Mycopathol. Mycol. Appl. **8**, 138—153.
4. ANDLEIGH, H. S. (1958) Investigations into the role of fungi in Pulmonary Diseases in India. Amer. Rev. Tuberculosis. **78**, 644—646.
5. DEEVE, D. L., BUTT, E. M. & HAMMACK, R. W. (1952) Recent Advances in clinical pathology 2nd Edition. Pp. 77 J. A. Churchill & Co. London.

ON A NEW GENUS OF THE ENDOMYCETACEAE

by

K. B. BOEDIJN

(22.X.1959)

INTRODUCTION

In the family Endomycetaceae up till now usually those yeast-like forms were placed, which are showing a more or less well developed mycelium. Furthermore all species grouped here are producing asci, which contain mostly only 4 spores, though other numbers may occur. Those asci are formed directly upon the mycelium and no real fructifications are ever developed.

We will see that this feature is of importance as is also the spore shape. The presence or absence of a mycelium however is immaterial.

When examining in Bogor, Indonesia, fruits of *Flacourtia inermis* thickly beset with *Mucor inaequisporus* DADE, I discovered between the growth of this mould inconspicuous yeast-like colonies, which proved to be of a representative of a new genus of the Endomycetacaeae. It will be described below. For the help with the latin diagnoses I wish to thank Dr H. C. D. DE WIT.

Endomycopsella BOEDIJN nov. genus.

Mycelium ramosum, septatum, bene formatum. Conidiophorae penicilliformes. Conidia cylindrica, hyalina. Rami verticillati (pseudomycelium disitur) in blastosporis disjunctis dispositi. Blastosporae in asci mutantes. Asci globosi, subglobosi vel elongati, 4 spori. Sporae conglobatae, saturniformes, tenuiter echinulatae, fuscae.

Mycelium branched and septated. Conidiophores with more or less penicillium-like branching. Conidia solitary, cylindric, hyaline. Pseudomycelium-like lateral branches originating from budding cells in whorls around the supporting hyphae. The majority of those cells transformed into asci, without any sexual act. Asci globose, subglobose, sometimes elongated, 4 spored. Spores conglobate, saturnine, delicately echinulate, brown colored.

Endomycopsella phaeospora BOEDIJN nov. spec.

Mycelium ramosum, septatum. Hyphae 3—14 μ crassae. Conidiophorae erectae, penicilliformes 85—340 μ longae, ad basim 3—4 μ ad apicem 4—10 μ crassae. Phialidae 27—48 × 3½—7 μ.

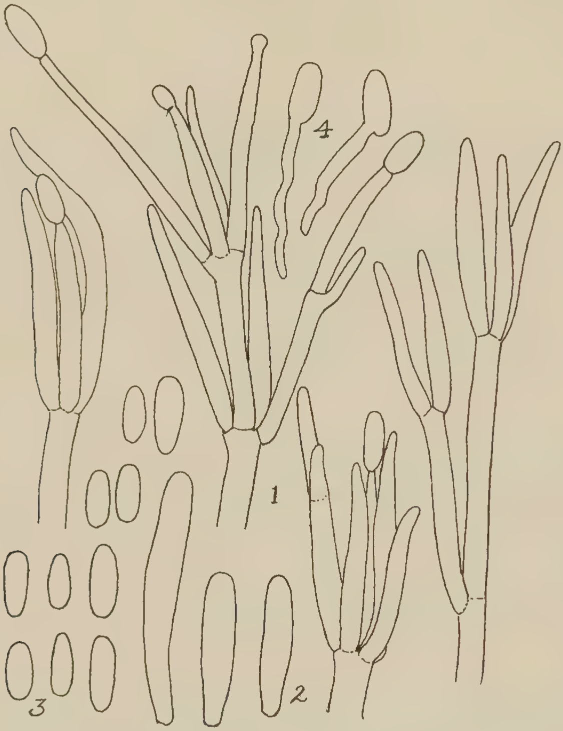


Fig. 1. *Endomycopsella phaeospora.*
1 Conidiophores. 2 shedded phialide-like branches. 3 conidia. 4 germinating conidia.

Conidia solitaria, hyalina, cylindrica 10—24 × 3½—7 μ. Pseudomycelium verticillatum, dispositum per blastosporas disjunctas. Blastosporae in asci mutantes. Asci 4 spori, globosi, subglobosi vel elongati, 14—20 μ diam. vel 24—44 × 7—14 μ. Sporae conglobatae, saturniformes, fuscae, tenuiter echinulatae, 10—14 × 7—8½ μ.

On the original substrate, colonies very inconspicuous, flat, smooth, glossy and of a dirty brown color. On Thaxter agar only slight growth. The colonies dendritic, flat and penetrating deeply into the agar. On rice agar some more growth. Colonies flat, inconspicuous and very thin. On togè agar growth good. Colonies nearly colorless or faintly tinged brownish, flat, the surface humid, glossy and rugulose. On oatmeal agar growth also good. Colonies deeply penetrating into the agar, flat, dirty brown, surface smooth, humid and glossy.

Mycelium richly developed, septated, sometimes constricted at the septa, 3—14 μ in diam. All cells uni-nucleate. Mainthreads at first unbranched over rather long distances, afterwards more or

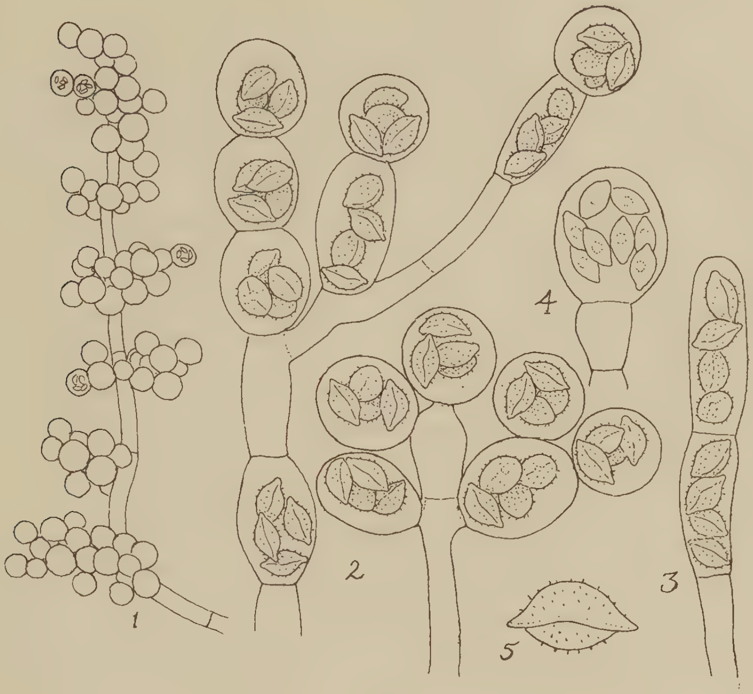


Fig. 2. *Endomycopsella phaeospora.*
1 Ascophorous hypha. 2 Parts of lateral branches with asci. 3 elongated asci. 4 large ascus with 8 immature spores. 5 ascospore.

less strongly branched. From the creeping mycelium ascending conidiophores 85—340 μ long are originating. They are at the place of attachement 3—4 μ, near the tops 4—10 μ broad. Here they are showing a penicilliform branching, which sometimes however is more or less irregular. At the top of the conidiophore a whorl of 3 till more phialide-like branches may originate, or the branching is 2 till 3 verticillate. The last phialide-like branches 27—48 × 3½—7 μ are bearing the conidia. They are mostly shed together with the conidia and may even germinate. Conidia solitary, colorless, cylindric with rounded poles or sometimes one pole slightly attenuated, 10—24 × 3½—7 μ, uni-nucleate. Germinating with a mycelium. After conidia formation or at the same time the mycelium is producing budding cells on the ends and the sides of long hyphae. Those cells singly or in unbranched or branched chains, are forming whorls around the supporting threads. They are for the most part transformed into asci without any sexual act. Asci mostly globose till subglobose, 4 spored 14—20 μ in diam. or 20½—24 × 15—19 μ. Spores conglobate. Often some of the elongated mycelial cells are also changed into asci, which are 24—44 × 7—14 μ and show the spores uniseriate.

Spores saturnine, the rim showing a weak bend on the sides, at first smooth and hyaline, at maturity delicately spinulose and brown colored, 10—14 × 7—8½ μ. Exceptionally a few large 8 spored asci were observed. In old cultures the asci are separating. Java, Bogor on fruits of *Flacourtia inermis*.

An undoubtedly similar plant was described by WELLS (4) as *Ascocybe Grovesii*. Here the mycelium forms separate conidiophores and the asci are united into heads at the top of special branches, called ascophores by the author of the genus. The asci are 4 spored and the ascospores are hat-shaped. In *Endomycopsella* we have also conidiophores and the asci arise on separate threads, but those are unspecialized and bear the asci along their whole length.

Other related genera are *Endomycopsis* and *Endomyces* and it is evident that *Ascocybe* as well as *Endomycopsella* have to be placed in the family Endomycetaceae.

One of the outstanding characters of this family, which was already pointed out by ZENDER (5) is to be found in the shape of the ascospores. These are provided with an equatorial ridge, or this rim is at the bass of the spore giving the appearance of a hat. The first type is known as saturnine, the second as cucullate ascospore. So all yeast-like forms which are showing such ascospores are to be placed in our family. We have then to add the genera *Ascoidea, Hansenula, Hanseniospora* and *Schwanniomyces*. The first is a somewhat aberrant genus, with multinucleate cells, but with true conidiophores, whereas the many-spored asci are containing hat-shaped ascospores. *Hansenula, Hanseniospora* and *Schwanniomyces* with typical saturnine or cucullate ascospores, but without mycelial formation are to be considered to represent reduced forms. Such a condition is not at all rare in many groups of the Fungi. The family Endomycetaceae will contain accordingly the following genera.

Endomycetaceae.

Ascoidea BREFELD Unters. Myk. 9.91.1891.
Ascocybe WELLS Mycologia 46.40.1954.
Endomycopsella BOEDIJN.
Endomycopsis DEKKER Verh. K. Akad. Wetensch. Amsterdam 2, 28.264.1931.
Endomyces REESS Bot. Unters. 77.1870.
Hansenula SYD. Ann. Mycol. 17.44.1919.
Hanseniospora ZIKES Centralbl. Bakt. etc. 2, 30.148.1911.
Schwanniomyces KLOECKER Centralbl. Bakt. etc. 2, 25.294.1909.

Literature

1. BEDFORD, C. L. (1942) A taxonomic study of the genus Hansenula. Mycologia **34,** 628—649.

2. Lodder, J. & Kreger-van Rij, N. J. W. The yeasts, a taxonomic study. 1952, xi & **713** pp. 268 fig.
3. Verona, O. & Zardetto de Toledo, O. (1959) Considerazioni intorno ad Endomycopsis fibuliger (Lindner) Dekker. Mycopathol. et Mycol. appl. **10,** 209—215, 5 fig.
4. Wells, D. E. (1954) Ascocybe, a new genus of lower Ascomycetes. Mycologia **46**, 37—51, 14 fig.
5. Zender, J. (1925) Sur la classification des Endomycétacées. Bull. Soc. Bot. Genève, 2e sér. **17,** 272—302.

LIBRI NOVI

BONNER, JOHN TYLER: The Cellular Slime Molds. Princeton University Press. Princeton, New Jersey. Investigations in the Biological Sciences, No. 1. 1959. pp. viii, 150. Price: $4.00

In the last 25 years, and especially since 1945, increasing interest has been shown in that group of organisms classed in the *Acrasiales*. These organisms are considered particularly useful in the study of developmental morphology. According to BONNER, the Mycotozoa (Myxomycetes) may be divided into four orders: *Myxomycetales* (Myxogastrales) or "true slime molds," *Plasmodiophorales* and *Labyrinthulales*, including two groups of plant parasites; and the *Acrasiales*, the "simple", "amoeboid", or "cellular slime molds",

After describing each of the orders of slime molds, the author suggests that the origin of the *Acrasiales* may be found in the free living amoebae of the soil. Three major points concerning the significance of aggregation in biology include: the aggregation process may serve as a partial substitute for sexuality; for some forms this may be the channel by which a truly multicellular condition has been achieved; and aggregation is an important tool in the study of the mechanisms of development.

A brief survey of the Acrasiales shows that the genera are placed in three families. *Sappinia* is placed in the Sappiniaceae, *Guttulina* and *Guttulinopsis* in the Guttulinaceae, and *Dictyostelium*, *Polysphondylium* and *Acytostelium* in the Dictyosteliaceae. *Acrasis* and *Coenonia* are known only from their original descriptions. A total of sixteen species is known in the order, of which those in the Dictyosteliaceae are used in most of the studies described in this book. Details of morphology are presented from spores and spore germination, through the "vegetative stage," the " aggregation stage", and the "migration stage", to the "culmination stage" of *Dictyostelium discoideum*. Other species descriptions are given in terms of variation from this basic pattern of the life cycle. Different stages are illustrated diagrammatically and by photographs. Macrocysts, microcysts, and spores are also illustrated. A hypothetical phylogenetic tree of the order is presented and discussed.

Since the book is designed as a survey and analysis of 123 papers or books published on this order and 42 supplementary to such a discussion, it is not surprising to find a minimum of graphic and tabular material and a maximum of discussion. Two principal problems are considered: the problem of morphogenetic movement, and variation and the problem of differentiation.

Three mechanisms of movement are considered. The substance acrasin, which has not yet been chemically defined, has been found to be important throughout these three stages: aggregation, migration, and culmination. As the scattered amoebae, which feed on bacteria, come together in the aggregation stage, a change comes about in the cell walls which permits cells to adhere to one another. Depending on the species, 24 to 2200 cells may be present in an aggregation. Still unknown are the factors which control aggregate size. Apparently not just one cell but any group of cells in the mass may possess whatever activator agent is required. Movements have been studied from many points of view and with numerous techniques: cell size, shape, and position within an aggregate; shuffling of portions of aggregates of clones, strains, and species; stains, lighting, chemical stimuli, variation in nutrients within an agar; and variations in cell orientation. Mechanics of movement are still a long way from being understood.

In spite of much experimentation on variation and methods of control of differentiation within an aggregate, there are still many unsolved problems. Variations are considered as between species and strains, between clones, and between the cells within a clone. Cell size, speed of movement, position within an aggregate in relation to tip or tail, and other qualities have been checked for genetic variation, but little information has yet come to light to indicate that each cell in an aggregate is not totipotent; that is, each cell in an aggregate appears to be genetically capable of performing any function at any stage in the life cycle that any other cell can perform.

The author is well known for his studies in this group of organisms. This fact is emphasized by the 13% of the citations by him or by him and with his students. The book is well written, easily read, and without noticeable typographical errors. It will serve as an excellent introduction to this poorly understood group of organisms for those unfamiliar with them, and a morphogenetic review for those in the field.

Wm. Bridge Cooke

Robert A. Taft Sanitary Engineering Center
Cincinnati, Ohio

Pierre Dansereau. *Biogeography: An Ecological Perspective.* New York, N.Y. The Ronald Press, 1957. 394 pp. $7.50.

As indicated in the title, this work approaches Ecology from a broad viewpoint including the interplay of plants, animals, and man with the environment. The great bulk of the discussion, however, is concerned with the vascular plants. The animal and microbiological viewpoint is merely mentioned in passing. It is not within the province of this review to evaluate the technical detail or accuracy of the theories and data presented. The author, himself, points out that the scope of the book only allows a review of the vast field covered. As such it will be invaluable to the beginning or advanced student of ecology.

The book is divided into five chapters or "levels" of discussion: Historical; Bioclimatological; Synecological; Autecological and "Mans Impact upon the Landscape". The text is well organised and written in an interesting style. The book is profusely illustrated with charts, tables, photographs and diagrammatic drawings which are original with the author. These features, together with an extensive glossary at the end, make the book a valuable source of information regarding the language, terminology and various theories of the field of ecology. The author, who has traveled widely and become personally intimate with many world-wide aspects of plant distribution is particularly competent to present such a broad approach.

Department of Botany
University of Michigan
Ann Arbor, Michigan

LEWIS E. WEHMEYER

ALLEN, ROY M.: *Photomicrography*, 441 pp., 235 figures, 54 plates in black and white and 1 color plate. D. Van Nostrand Co., Inc. Princeton, N. J. Second edition November 1958, reprinted February 1959.

Long and authoritative guide to the making of photographs with a microscope, this useful volume now encompasses new designs of microscopes, cameras, and accessories, new methods and applications that present an up-to-date manual of practical modern usage. Included are such important subjects as phase and interference microscopy, electron microscopy, uses of color photography, and the application of polarized light.

Presentation of photographic equipment in all its details (cameras, optical systems, light sources) is exhaustive. Highly important are the illustrations concerning common errors and failures in photomicrography (uneven illumination, not axial lighting, excessive diffraction, etc.). The plates are excellent and most instructive, they bring examples from the whole field of microbiology. Although the three-dimensional presentation of the subject is highly important in botany, this manual proves that even with the two-dimensional photomicrography one can get excellent results from botanical specimens, let alone, from the practically two-dimensional objects in mycology. Drawings from mycological specimens are instructive, however, photomicrographs are documentary evidences. This manual can be warmly recommended to microbiologists in general and to mycologists in special.

TIBOR BENEDEK

Fungus Diseases and Their Treatment, edited by R. W. Riddel and G. T. Stewart, XVII, 256 pp., 145 figures. Butterworth & Co. Ltd. London, 1958. Price 45/- net.

This book is a condensation of papers read at the SYMPOSIUM ON FUNGUS DISEASES held in London during 1958. The SYMPOSIUM was designed to bring together individuals with different approaches to each topic, and was characterized by contrasts in subject-matter. Among the 34 contributors, the brunt of the work was done by British investigators, yet, French, American and Belgian research was well represented. This SYMPOSIUM contains two parts: (1) Pathology, Clinical Features, and Epidemiology; (2) Treatment. The first part contains 22 papers, and the second one 9 papers. A number of papers were dealing with superficial mycoses, with special attention to infections caused by *Tr. rubrum*. Moniliasis, Aspergillosis, Histoplasmosis are covered clinically, pathologically and immunobiologically. The Treatment covered older and newer methods, with special reference to antifungal drugs.

The almost proverbial conformity of opinion of the dermatomycological confraternity came again to the fore in this SYMPOSIUM. I refer particularly to the chapter of Calnan on "*Trichophyton rubrum* infection" and to that of Holmes on "Tinea pedis in miners". These chapters excel in poor methodological knowledge and a complete lack of knowledge of the pertaining literature. These authors still believe that a positive fungal culture is at the same time a "fungus disease". For Calnan *T. rubrum* infection of the palms and soles is equally resistant whether the nails are involved or not . . . "Nail infection is at present virtually incurable .." As far as these "infections" are considered as "fungus infections" and treated as such, they will be "resistent" for the simple reason because they are not "fungus" infections. What he describes as fungus infection of palms and soles and the nails is pompholyx of the palms and soles and of the nail organs, a condition not of fungal origin, but endoparasitic-bacterial. With this correct etiological diagnosis the "cure" is straight and simple. He does not mention whether he ever observed the primary lesion of these hand- feet- and nail mycoses. There exists a rich literature on this topic which went completely unnoticed by this author. The presentation of Holmes moves on the same level as the question stood in 1911 when Malcolm Morris "assumed that white soggy skin between the toes was evidence of tinea pedis" . . . He repeats the same stock errors by sheer conformism which is slowly overcome in the States. Knowledge of this literature would have helped to arouse some doubt about the etiologic role of fungi he was able to culture. His list is an outstanding counterpart of the list of fungi which mycologists prepared not so long ago about the two dozens or so of fungi as "causative agent" of pinta!! Mycologists became the victim of their ignorane of the primary lesion in pinta which HERREJON proved to be a spirochaetosis.

The other chapters of the SYMPOSIUM do not carry any problematic. Their material was adequately handled by the respective author. They are well written, well presented and adequately illustrated. Every paper brings selected references. The therapeutic part of the Symposium gives an excellent and objective presentation and orientation about the newer antifungal antibiotics. All in all, this SYMPOSIUM gives a good survey about the present trend and current of medical mycology. It is a useful publication for dermatologists and mycologists working in this borderline field. The lesson derived for both groups of workers is that conformism is a fiend of progress in any field of science. They finally have to give up the age old error that a fungus culture, no matter whether pathogen or non-pathogen, is a fungus disease. They have to increase their methodologic knowledge and they have to know the foremost basic rule of investigation in medical mycology that is the knowledge: the etiology and pathogenesis of the primary lesion in any condition. The production of the book (paper, printing, illustrative material) is satisfactory.

TIBOR BENEDEK

WYBURN-MASON, ROGER: The Reticulo-endothelial System in Growth and Tumour Formation. London, Henry Kimpton, 1958. 206 pp., with 79 illustrations in black and white. Price 42/- net.

The protective function of the reticulo-endothelial system with its ability to extract foreign substances, bacteria and cells and effete red and white blood corpuscles from the circulation is well known. In this book the author has thought to show that this is only part of the function of the cells of this system. They appear to extract from the circulation and concentrate the various physiological substances, including proteins, amino-acids, fats, lipids, sterols, vitamins and hormones, and to bring them into contact with the parenchymatous, epithelial, muscle and nerve cells and to receive from these same cells their metabolic products, in so doing playing a part in cellular excretion.

The text is well written, concise and clear. The illustrations are well chosen. The production of the book (paper, printing) is excellent. Medical mycologists, beside internists and general pathologists, have to be interested in this monographic text, because the reticulo-endothelial system plays also a role of basic importance in the course of the deep-seated mycoses. References are adequate. However, the reviewer regrets the "blind" references, that is references without title of the papers. In a monograph there is no excuse for this omission. A detailed index closes the work. It can be generally recommended.

TIBOR BENEDEK

WAKSMAN, S. A.: *The Actinoycetes Vol.* 1. *Nature, Occurrence, and Activities.* The Williams and Wilkins Company, Baltimore, 1959. pp, xi, 327, 107 Fig., 77 Tables, Price: $12.50.

This is the definitive work on the Actinomycetes. It is written by the man whose name is synonymous with research in this group of organisms. Two more volumes are promised in this series and are referred to constantly throughout this one. A preface, in which the author indicates his own personal interest in the group, and an introductory in which general milestones in the study of the Actinomycetes are listed, together with certain typographical conventions to be made in the book, precede the main presentation which is divided into 18 chapters. The importance of the study of this group of organisms is attested to by the facts that nearly 25 antibiotics derived from Actinomycetes are in general use today, and that two-thirds of the 2.4 million pounds of antibiotics which were produced in the United States in 1955 alone, at an estimated value of more than a half billion dollars, were derived from the Actinomycetes.

Among the topics which are considered in this general volume are included: history, methods of isolation, identification, cultivation and preservation; distribution in nature; nomenclature and general systems of classification; morphology, cytology and life cycles; variations, mutations and adaptations; physiology, including growth and nutrition, mineral metabolism, biochemical activities and lytic mechanisms; the production of enzymes, vitamins, other growth-promoting substances and pigments; antagonistic properties; the production of antibiotics; the role of the Actinomycetes in the decomposition of plant and animal residues; and the diseases of plants and animals caused by these organisms.

Dr. WAKSMAN is strongly in favor of considering these organisms bacteria. He presents 10 strong arguments in favor of this proposition while he could only find four arguments, several of which are very weak, indicating their relationships with fungi.

Approximately 1200 references are cited throughout the text and listed in a comprehensive bibliography. Of these, 6% were written by the author. In a treatise such as this, in which an attempt is made to summarize all the literature, certain items are bound to be left out. This is partly because an item was considered unimportant or of no significance, partly because a report was not known to the author. To those for whom this book may appear, or may be assumed, to be a complete encyclopedia of information about the Actinomycetes the statement should be made that in both his preface, his introductory, and his epilogue, Dr. WAKSMAN admits that first consideration was given to his own work, and the work of his students. Throughout the text it is apparent that in addition to this work, that published in Russia has been given a prominent place in the treatment. This is good because otherwise much significant work could easily be overlooked partly because of availability of references, partly because of the language barrier. Thirdly, the works of other students of these organisms in the United States, England, Japan, and many other countries throughout the world are included.

It is interesting that in his introduction of the term "colony" in the text, Dr. WAKSMAN indicates that the actinomycete colony arising from a single spore is not a true colony but a single organism. On page 250 the term "created" is used in reference to recognition of genera of thermophilic organisms. One wonders if this was meant in the Lloydian sense or if the author accepts these genera and really should have used the word "recognized".

At several points in the text the decomposition of lignin is considered. On page 247 this is given a half column of space. However, whenever this is mentioned no references are made to research on the subject although Table 68 on page 244 gives a clue to research that has been done on this problem. Additional information in this field would be of much help. In relation to composting, only work with green and stable manures is considered. No reference was found to work which has been done on the composting of garbage and and municipal refuse as a means of disposal of a troublesome refuse and the conservation of an important resource.

The chapter on morphology, cytology and life cycles is primarily concerned with morphology. A brief section is given over to sexuality and another to staining properties. It would seem to this reviewer that in this connection a lot more space could have been given to cytology, the demonstration of nuclear materials or artifacts assumed to be nuclear bodies in cells, and other matters more readily demonstrated by the electron microscope. Electron micrographs are used as illustrative materials in various places in the book but either have been used insufficiently or are not available for use in connection with the development of the topic of cytology. In the section on sexuality, the work reported in nine references is summarized in four paragraphs. Figure 36, p. 75, reproduces a life cycle proposed in a paper which is only cited as a reference to which the reader may turn. Upon turning to this reference one wonders why Dr. WAKSMAN did not choose to consider it point by point in his text, or if not worthy of that treatment, why he used it at all.

It seems to this reviewer that if the material on heterocaryosis, presented in Chapter 6 on variation, mutations and adaptations, is all that is available on this subject in relation to the Actinomycetes, here is a very fertile field for work. In reading through various parts of the text it seemed that a number of phenomena which were described could well have been studied with this in mind.

The book is well written and easily read. Format and type are similar to those used in "Applied Microbiology," that is, two-column pages of folio size. Illustrative material is well chosen and placed in the text near the point of reference. Only two minor typographical errors were noted. The author and his publishers are to be congratulated on the production of a treatise which will be of value to anyone whose work embraces the Actinomycetes.

Wm. Bridge Cooke

Robert A. Taft Sanitary Engineering Center
Cincinnati, Chio

Gäumann, E. A.: *The Fungi*. A description of their morphological features and evolutionary development. Translated from the German by Frederick Lyle Wynd. Hafner Publishing Company, New York-London, 1952. Price $ 10.— 420 pp., with 440 figures.

This book describes the morphological and developmental features of Fungi. It especially emphasizes the natural relationship existing within this vast and intricate group of plants. Gäumann-Wynd have directed their efforts toward aiding the student to understand the Fungi rather than to classify them. The reader continually is aware that the natural phenomena presented by the Fungi are more important than rigid and artificial systems of classification. Taxonomy is regarded as a useful but flexible aid for the comprehension of major biological problems.

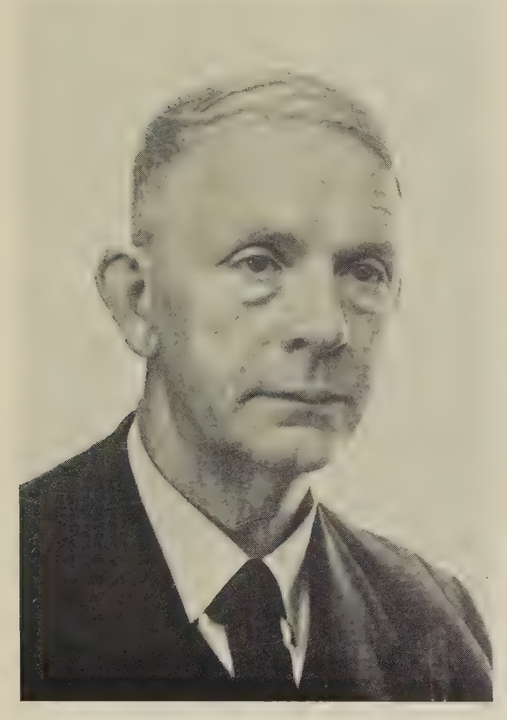
The Fungi are grouped in four classes: Archimycetes, Phycomycetes, Ascomycetes and Basidiomycetes. An appendix deals with the Fungi imperfecti. The illustrations are excellent and most instructive. The translation is highly satisfactory. The production of the book (paper, printing, illustrations) is impeccable. This work of Gäumann, like his other works on Fungi, does not need any further recommendation. They speak for themselves. For the students of general Mycology it is simply a "must".

Tibor Benedek

Bassi, Agostino: *Del Mal Del Segno*, translated by P. J. Yarrow. Edited, and with an introduction, by G. C. Ainsworth and P. J. Yarrow. Phytopathological Classics, No. 10. Published by the American Phytopathological Society, Ithaca, N.Y. XIII, 49 pp., Frontispiece: Portrait of Agostino Bassi.

Phytopathological Classics embraces titles that are unavailable in most libraries and provides translations of papers originally published in foreign languages. This publication brings the work of Bassi on Muscardine or Silk Worm Disease in excellent English translation. The original was published at Lodi (Italy) in 1835. Bassi's name survived because he was the first to elucidate the nature of a microbial disease of animals when he proved the fungal nature of the muscardine disease of silkworms. This treatise like the other numbers of this series of Phytopathological Classics are of interest to pathologists, mycologists, plant breeders, physiologists, and historians of science.

Tibor Benedek



I. JØRSTAD

Botanical Museum, Oslo, Norway.

Gallery of Contemporary Noted Mycologists

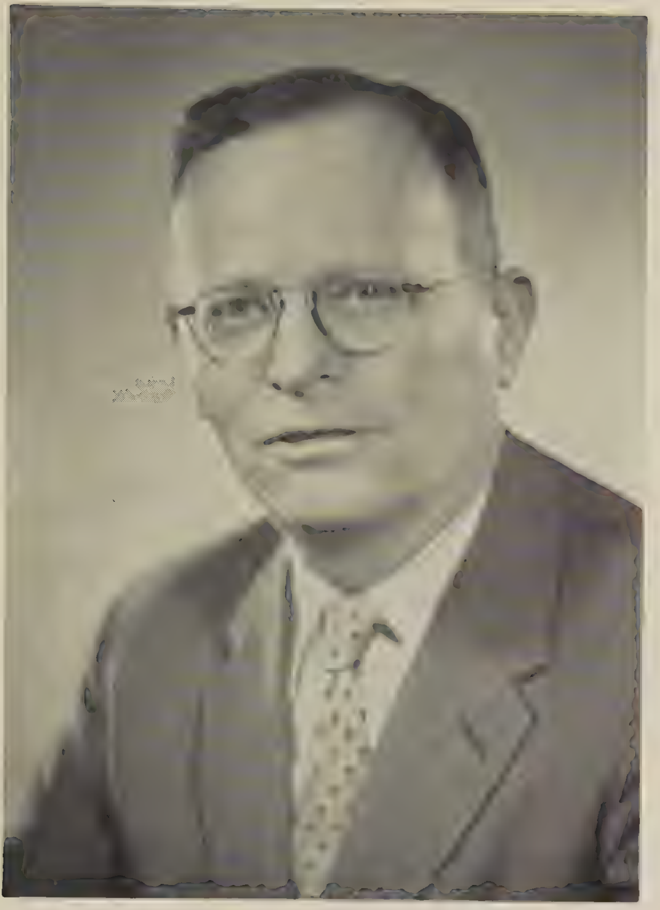
65

T. DOMINIK

Forest Microbiological Laboratory
Forest Research Institute, Szczecin, Poland

Gallery of Contemporary Noted Mycologists

67



R. SINGER

Instituto Miguel Lillo
Tucumán, Argentina

Gallery of Contemporary Noted Mycologists

68

IV. VENEZUELAN CONGRESS OF PHTHISIOLOGY AND PNEUMOLOGY. DEEP MYCOSES IN VENEZUELA WITH SPECIAL REFERENCE TO PULMONARY MYCOSES.

The "IV Congreso Venezolano de Tisiologia y Neumologia" with an inclusion of a SYMPOSIUM ON MYCOSES took place at Valencia, Venezuela, December 8—12, 1959.

IV CONGRESO VENEZOLANO
DE TISIOLOGIA Y
NEUMONOLOGIA

VALENCIA - 8 al 12 de Diciembre de 1959

Fig. 1. Emblem of the Congress.

Venezuela, birthplace of the great South American liberator, Simon Bolivar, is a land of great variety. Snow-covered peaks, lush tropics, rolling plains and jungle make up the terrain. A costal range rises to 5,000 feet, and in the West, terminating at Barquisimeto, is the Andean Chain with a number of peaks over 15,000 feet high. Back of the Andes lie the rolling treeless plains (llanos) of the Orinoco river basin, and South of these (making up almost one-half of the territory of Venezuela), are the vast forested, unexplored Guayana Highlands. In the northwest, bordering the Carribean, is the oil-rich Lake Maracaibo region, tropical and hot.

Venezuela is the Northernmost country of South America, bounded by the Carribean Sea on the north, Colombia on the west, British Guiana and Brazil on the east, and Brazil on the south. Its vast territory covers over 350,000 square miles.

These complex geographical features make this country a hotbed — El Dorado — of fungus diseases, superficial and systemic. Fungus diseases of the lungs, beside the wide spread tuberculosis, represent one of the foremost problems of Public Health.

Due to the unfaltering endeavor of Prof. Dr. José Ignacio Baldó a unique forum was created for the study of mycoses in Venezuela: COORDINATING COMMITTEE FOR A NATION-WIDE STUDY OF MYCOSES (Comisión Cordinadora del Estudio Nacional de las Micosis). Prof. Baldó is the president of this Committee.

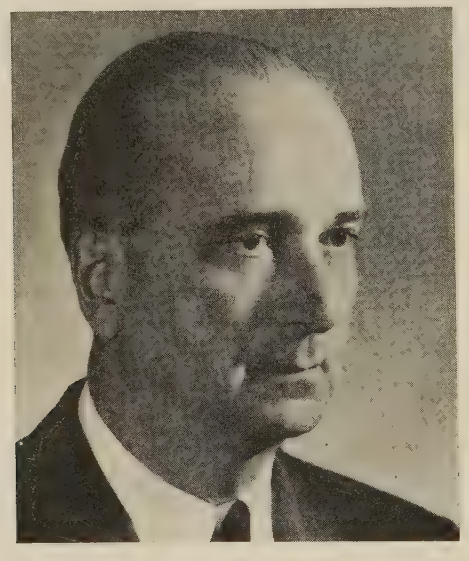


Fig. 2. Prof. Dr. José Ignacio Baldó
Honorary President, Sociedad Venezolana de Tisiologia y Neumologia. President, Coordinating Committee for a Nation-wide Study of Mycoses.

Since the organization and the function of this highly important Committee and its achievement during the relatively short duration of its existence are hardly known in other parts of the world, it seems to be worthwhile to give a short presentation of its organization, its aims, and its achievements for the central coordination of the study of mycoses.

The Coordinating Committee for a Nation-wide Study of Mycoses [1]).

The purpose of this Committee is to coordinate all of the nation's services, both private and governmental, that devote part or all of their efforts to the study of mycoses. The Committee is composed of thirteen persons specialized in the various phases to be covered by the study (clinicians, mycologists, bacteriologists, parasitologists, dermatologists, epidemiologists, otorhinolaryngologists, and odontologists). The Committee is an autonomous agency with headquarters at Caracas, and has as its purpose the coordination of all activities tending to promote, organize, and carry out the study of mycoses in Venezuela. The Committee has a broad field of endeavor. It seeks to establish contact among those interested in mycological problems, it requests and provides consultant services, it files national clinical reports, and it plans the establishment of a National Mycology Archives. Furthermore, it helps bring about standardization in research techniques; it will furnish antigens and offer the diagnostic facilities of specialized laboratories; it plans studies of various types in accordance with data received; it drafts the standards to be followed in such studies by participating individuals and institutions.

---

[1]) Cf. Baldó, J. I.: El estudio de las micosis en escala nacional, Acta Medica Venezolana, vol. 5, no. 4, Julio-Agosto. Those who wish further information on details, address: Instituto Nacional de Tuberculosis, El Algodonal-Antimano, Caracas, Venezuela.

In the relatively short period of time in which the Committee has been in operation in Venezuela, it has produced results which are most encouraging. They include the following:

1. The establishment of a Central Registry for cases of mycoses. It was designated as a central office, located in the capital city Caracas, and its function is to be that of collecting and presenting, for statistical analysis and interpretation, all of the informations contained in the clinical histories prepared by special subcommittees.
2. Plans have been made for the establishment of subcommittees which, in due course, will study the problem of the nomenclature and treatment of mycoses.
3. Agreement was reached on the establishment of a bibliographic bulletin, to contain reviews on books and periodicals.
4. Standards were drawn up to govern the procedure for, and the interpretation of, intradermal tests.
5. A plan presented by the Tuberculosis Division of the Ministry of Health and Social Welfare was already carried out, consisting of cutaneous testing with histoplasmin, coccidioidin, tuberculin and radiological survey of the town Cumanacoa, State of Sucre. Nearly 10,000 persons were covered. This survey was completed in three months. This special endeavor has the purpose of preparing an epidemiological map of mycoses in Venezuela.

This survey, the Committee recognizes the fact with deep gratitude, was carried out with the generous and valuable cooperation of distinguished scientists from the United States, among others, of Dr FORREST W. CROSS (Chief, Biologics Preparation Laboratory, Tuberculosis Program, Division of Special Health Services, Department of Health, Education and Welfare, Washington, D.C.) who furnished all the histoplasmin used in the survey and in subsequent research; of Dr. CHARLES E. SMITH (University of California School of Public Health,) who supplied all the coccidioidin used in these programs; and of Dr. LIBERO AJELLO (Chief, in charge of Mycology Unit, Communicable Disease Center, PHS, Department of Health, Education and Welfare, Washington, D.C.) who cooperated in the serologic testing of cases in the survey.

6. Thanks to the Committee's initiative, a modern serological laboratory has been installed in the Institute of Tropical Medicine, Central University Caracas, where satisfactory results have already been obtained in the preparation of antigens of the yeast-like phase of *Paracoccidioides brasiliensis*, *Blastomyces dermatitidis*, *Histoplasma capsulatum*. Moreover, the technique for the complement fixation has been established.

7. A plan has been drawn up to furnish the national universities (Venezuela has now five universities, with the oldest one, the Central University in Caracas, established in 1722 by the Spanish king) with photographic material for teaching purposes, to show the principal mycoses in their different aspects with which the instruction will deal. A brief written text is attached to each film and the running time of a film on a particular mycosis will be 45 minutes.

8. A proposal has been drawn up for a National Mycoses Archives the regulations of which foresee its operation under the direction of a Technical Council, which will set the policy for its relations with institutions and individuals both national and foreign.

9. In order to summarize simply and clearly the basic information on the principal mycoses, the Committee has issued a booklet, entitled "Cartilla Mycologia" (Mycological Primer), which contains a brief description of each of the mycoses, with stress being placed on definition, epidemiology, clinical aspect, diagnosis and treatment. The booklet is especially directed to the medical profession and to students in the last two years of the medical sciences.

Due to the unrelenting effort, driving strength and organisatory genius of Doctor BALDÓ the Government of Venezuela presented to the XI Meeting of the Directing Council of the Pan American Health Organization, for its consideration, the following draft resolution:

1. To recommend to the Member Countries of the Pan American Health Organization that they promote the creation of Coordinating Committees of nation-wide scope for the study of mycoses;
2. To recommend to the Member Countries of the Pan American Health Organization the undertaking of epidemiological surveys as a means of ascertaining the magnitude of the problem of mycoses in the Americas.

THE SYMPOSIUM ON MYCOSES

Backed by this unique, scientific organization it is no wonder that the SYMPOSIUM ON MYCOSES became a complete success. The Committee arranged jointly with the IV Venezuelan Congress of

Phthisiology and Pneumology a symposium on "DEEP MYCOSES IN VENEZUELA WITH SPECIAL REFERENCE TO PULMONARY MYCOSES", with the participation of Venezuelan specialists and especially invited experts from abroad. The first two days of the meeting were consacrated to the problems of tuberculosis of the lungs, while the last three days dealt exclusively with the deep seated, systemic and pulmonary mycoses.

Altogether 33 papers have been presented on histoplasmosis, coccidioidomycosis, paracoccidioidomycosis, criptococcosis, chromoblastomycosis, maduromycosis, sporotrichosis, rhinosporidiosis; on mycoses of the lungs due to *Aspergillus* sp., *Mucor* sp., and *Penicillium* sp. The papers presented were of the highest scientific quality as to methodological knowledge, clarity of presentation, precision of the topics outlined, with the additional outstanding illustrative material concerning clinic, pathology, epidemiology, mycology, in slides, graphs and charts. It is no room here to discuss the outstanding features of the individual papers. In due time, they will be presented in this Journal and they will speak for themselves.

COLEGIO
DE
MEDICOS

Fig. 3. Home of the Medical Society of Valencia, where the sessions of the Congress were held.

The Congress which held its session in the auditorium of the Colegio de Médicos, Home of the Medical Society of Valencia, was opened December 8th, by Dr. ARNOLDO GABALDÓN, Minister of Hygiene and Social Assistance of Venezuela, with an inaugural address. Then, Dr. FRANCISCO MELET, Governor of the State of Carabobo, extended the welcome of his Government to the audience

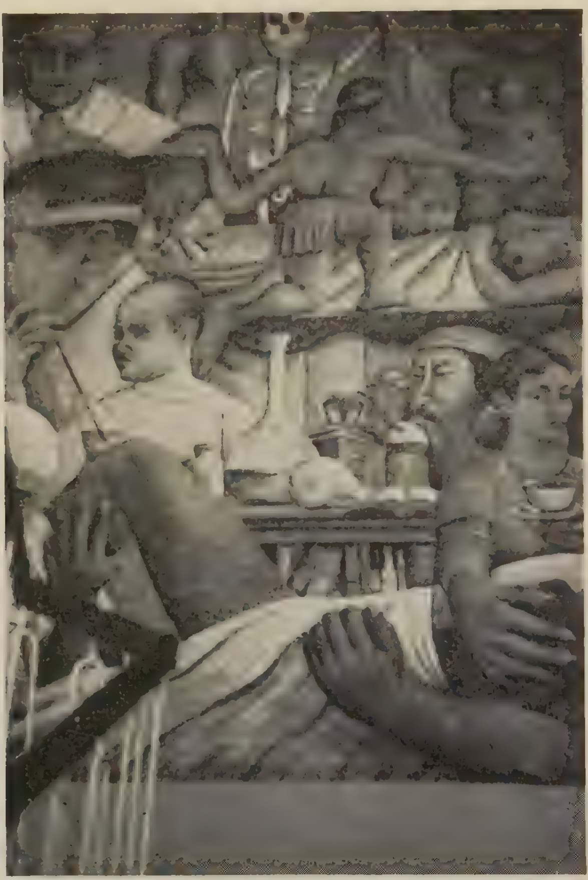


Fig. 4. "Activity of the Physician".
Detail from a Fresco in the Colégio de Médicos.

(about 650 members of the Society of Tuberculosis were present). After the words of the President of the Venezuelan Society for Phthisiology and Pneumology, Dr. César Rodriguez, the President of the Congress, Dr. Carlos Ortega Gragirena closed the inaugural session.

Following is the list of the invited scientists from abroad in alphabetical order:

Libero Ajello, Communicable Disease Center, Mycology Unit, Chamblee, Georgia, U.S.A.; Floriano P. de Almeida, Universidad de Saõ Paulo, Brasil; Tibor Benedek, Stritch School of Medicine, Loyola University, Chicago, Ill. U.S.A.; Phyllis Q. Edwards, Tuberculosis Program, U.S. Public Health Service, Washington 25, D.C. U.S.A.; Michael L. Furcolow, Chief, Kansas City Field Station, USPHS, Kansas City 3, Kansas, U.S.A.; A. Gonzáles

OCHOA, Laboratorio de Micología, Instituto de Salubridad y Enfermedades Tropicales, Mexico 17, D.F. Mexico; ESMOND LONG, The Henry Phipps Institute, Philadelphia, Pa. U.S.A.; JUAN E. MACKINNON, Instituto de Higiene de Montevideo and President of the International Society of Human and Animal Mycology, Montevideo, Uruguay; PABLO NEGRONI, Faculdad de Ciencias Médicas, Centro de Micología, Buenos Aires, Argentina; G. SEGRETAIN, Service de Mycologie et Physiologie Végétale, Institut Pasteur, Paris, France; HENRY C. SWEANY, Missouri State Sanitarium, Mount Vernon, Missouri, U.S.A.

After the papers on SYSTEMIC MYCOSES OF THE LUNGS had been read, Friday afternoon, an outstanding discussion took place. The moderators were Drs. BALDÓ and LONG who monitored the session with the greatest tact, circumspection and efficiency. All the invited guests from abroad participated in the discussion which added some fine points to the papers read. However, in all sincerity, the consensus of opinion of the guest scientists was, that we could learn more from, than contribute to the outstanding papers.

There was only one scientific exhibit, one on HISTOPATHOLOGY OF THE PULMONARY MYCOSES, presented by Dr. ALBERTO ANGULO ORTEGA, director of the Department of Pathology, Instituto Nacional de Tuberculosis. The exhibit encompassed about 100 colored prints, 8 × 10″, of the finest quality as to the photographic technique and of superb presentation of histopathologic sections from tissues and fungi. Dr. ANGULO ORTEGA has to be highly complimented for this unique achievement.

Last but not least, it should be mentioned that coincidental with the Congress the SOCIEDAD VENEZOLANA DE TISIOLOGIA Y NEUMOLOGIA started a new journal REVISTA DE TISIOLOGIA Y NEUMOLOGIA, Dr. NESTOR L. RINCON, as the editor. Judged by the excellent papers contained in this first issue, it is hoped that this journal will take a lead in the field of tuberculosis, mycoses and allied diseases of the lungs, not only in Venezuela, but also in Latin America as a whole. A leading article gave homage to Prof. JOSÉ IGNACIO BALDÓ whose indefatiguable work in the organization of the fight against tuberculosis and allied diseases of the lungs was properly put in focus.

Beside the hard and most intensive work in the scientific sessions social amenities were not lacking. The tone was set by superb hospitality, genuine cordiality of our hosts. I would like to underscore two social events. Thursday, December 10th, the Governor of the State of Carabobo (the capital city is Valencia where the meeting took place) was the host to the convention at the BATTLE FIELD OF CARABOBO where the independence of Venezuela from Spain was definitely won, in 1821, under the leadership of the great liberator, Simon Bolivar. This is at present a public recreational park in the center of which the beautiful monument of independence

Fig. 5. The Monument of Independence at the Battle Field of Carabobo at nightly illumination.

fills the spectator with great awe. At advanced hour national dishes were served in the beautiful tropical setting.

The other interesting event was offered to the members of the Congress and their ladies by the Ministry of Labor and the Government of the State of Carabobo, Friday night, in the old Spanish, charming municipal theater. The program was "VENEZUELAN DANCES". The audience was most grateful for the opportunity, to take a glance at Venezuelan folklore in dance and song.

Our Venezuelan hosts can be proud of the great success of this Congress which revealed to the invited guests the highest standards of research and great talent of organization in present day Venezuela.

TIBOR BENEDEK

DA OCORRÊNCIA DE HANSENIASPORA VALBYENSIS KLOECKER EM APÊNDICE CECAL HUMANO E LESÕES EPIDÉRMICAS

A. Chaves Batista, R. Pessôa Coêlho & J. Rêgo Vieira

(30.VII.1959)

Instituto de Micologia — Universidade do Recife

Publicação no 183

O gênero *Hanseniaspora* foi criado por Zikes (Cbl. Bakt. Parasitenk. Abt. II, 30: 145, 1911) para comportar leveduras ascosporadas, de fórma apiculada.

No decorrer de várias décadas esteve em foco a questão da existência, fórma e número de esporos de algumas leveduras apiculadas, descritas por autores como Niehaus & Dvornik, havendo mesmo sido proposto pelo primeiro dêsses autores o novo gênero *Kloeckeraspora* (Zbl. Bakt. Parasitenk. Abt. II,86: 253, 1932 e 87: 97, 1932.)

Entretanto, Lodder & Kreger van Rij., em sua sistematização dos fungos leveduriformes (The Yeasts, North-Holland Publishing Company — 1952), firmaram a diagnose do gênero *Hanseniaspora,* admitindo a possibilidade de variação da quantidade de ascosporos por asco, desde que a continuidade dos cultivos, nesse fungo, induz a reprodução assexuada. O gênero foi definido, então, como caracterizado por "células apiculadas, à maneira de limão, ovais ou longo-ovais; reprodução por brotação bipolar; pseudo micélio ausente; ascosporos inicialmente esféricos, depois a modo de chapéu, um a quatro por asco; capacidade fermentativa comprovada; nitrato não assimilado."

Ao mesmo tempo estabeleceram Lodder & van Rij, para êsse gênero, uma única espécie — *H. valbyensis* Kloecker, que se tornou tipo.

Estudos mais recentes de Miller (Yeasts, News Letter, VI, I: 14, 1957) incluiram, nêsse mesmo gênero *Hanseniaspora,* mais duas espécies: *H. uvarum* (Nieh.) Miller e *H. osmophila* (Nieh.) Miller.

Revendo a literatura médico-micológica, encontramos o assinalamento de apenas um caso de *Hanseniaspora* sôbre o homem, como provável agente de uma onicomicose (Pijper, A. — Verslag Gewone Vergader. Koninkl. Akad. Wetenschap. Afd. Natuurkunde, 37: 868, 1928).

Os nossos achados desta espécie, sôbre apêndice cecal e sôbre lesões epidérmicas, ampliam, sobremaneira, o significado da patogenicidade provável de *H. valbyensis*, enquanto elastecem o conhecimento do "habitat" dêsse fungo.

A amostra que obtivemos de conteúdo de apêndice, do paciente M.N.L.L., foi-nos enviada pelo Dr. S. D. de Oliveira, imediatamente após a apendicectomia.

O enfêrmo sofria de apendicite crônica.

Pequenas porções dessa amostra foram plantadas no meio de Sabouraud-dextrose-agar, com acromicina, afim de inibir o desenvolvimento de bactérias. Após 3 dias, apareceram pequenas colônias leveduriformes, que foram isoladas e transferidas para placas de Petri, contendo malte-agar. Realizadas as observações morfológicas necessárias à sua identificação, logramos a identificação dêsse microorganismo como *Hanseniaspora valbyensis* Kloecker, cujas caracteristicas são as sequintes:

Crescimento em extrato de malte- aos 3 dias e à 30° C: células globosas (pouco frequentes) ou ovais, com os polos apiculados ou em forma de botelha, 2—8 × 2—4,5 μ; células longo-ovais e fusiformes, com as extremidades frequentemente alongadas, 4,5—10,5 × 2—4,5 μ; brotação simples e múltipla, uni ou bipolar, havendo, quando simples, uma base larga.

Observamos, ainda, células alongadas, formando curtas cadeias. Ausência de micélio e de pseudomicélio, bem como de anel e película.

Cultura estriada, sôbre malte-agar, aos 10 dias e a 30° C.

Microscòpicamente, as células têm as mesmas caracteristicas e dimensões das observadas sôbre extrato de malte; macroscòpicamente, as colônias são cremosas, cinza, brilhantes, lisas, planas, de bordos ondulados, nítidos, sem filamentização.

Cultura sôbre lâmina de batata-agar, aos 4 dias e a 30° C: ausência de pseudomicélio e micélio: desenvolvem-se curtas cadeias de células blastospóricas.

ESPORULACÃO

Sôbre agar de Gorodkowa, aos 10 dias e a 30° C.: ascos ovais, ovoides de extremidades apiculadas ou fusiformes, 4—9 × 2,5—4 μ, Fig. 1. Ascosporos globosos, ovais e em fórma de chapéu, 1 a 4 por asco, medindo, no máximo, 1 μ de diam. Em geral, os ascosporos dispõem-se aos pares ou em agrupamentos de 4, cujo contôrno é oval; a superfície de contacto dos ascosporos forma uma figura correspondente à letra "H".

Fermentação:		Assimilação:	
Glucose	+	Glucose	+
Galactose	—	Galactose	—
Sacarose	—	Sacarose	—
Maltose	—	Maltose	—
Lactose	—	Lactose	—

Nitrato de potássio — negativa.

Hanseniaspora Valbyensis Kloecker

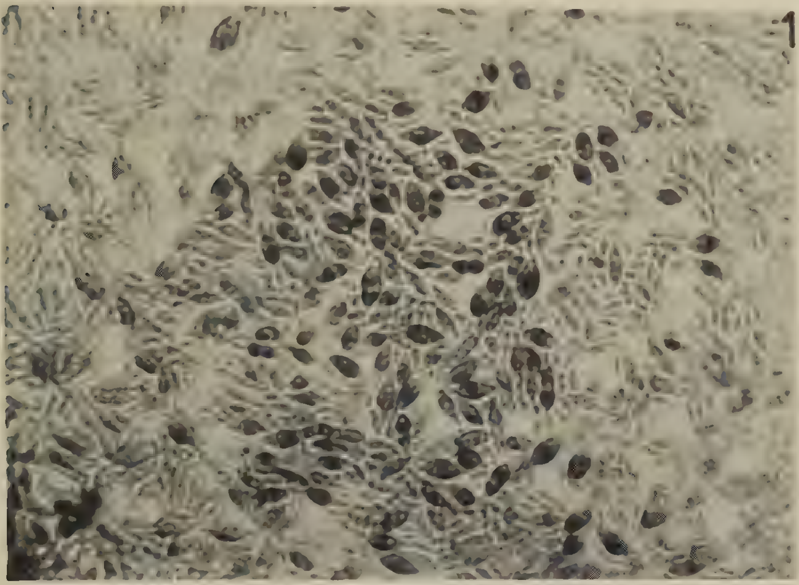


Fig. 1. Ascos, formados sobre o meio de Gorodkowa; cepa oriunda de apendice cecal.

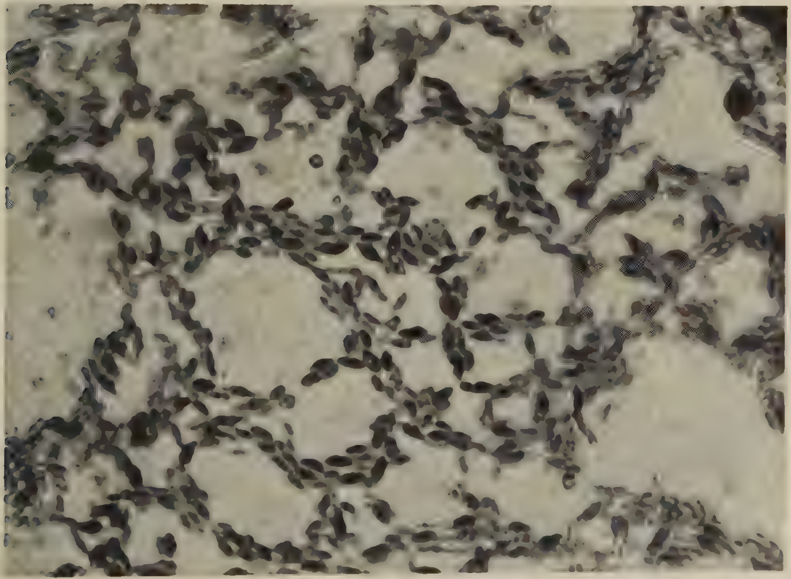


Fig. 2. Ascos de cepa proveniente de lesão epidérmica.

Esta nossa cepa apresenta-se sem exigências, quanto a fatores de crescimento e esporula bem, aos 10 dias, produzindo numerosos ascos, sob partenogênese, aparentemente.

Tem o no 1248, da Micoteca do I.M.U.R.

Outra amostra, sob no 2301, I.M.U.R., proveniente de lesão epidérmica de I.A.A., do sexo feminino, branca, 30 anos, doméstica, ensejou-nos, igualmente, o isolamento dessa mesma espécie de *Hanseniaspora* de que ora tratamos, também com produção fácil de ascos, no meio de Gorodkowa, Fig 2.

Neste caso a levedura em tela achava-se associada, a princípio, a uma lesão interdigital, da mão esquerda, que começara há cerca de um ano e progredira na direção do punho, com a formação inicial de pequenas vesículas pruriginosas, que se transformaram em pústulas, acompanhadas de descamação furfurácea da epidérme. A enfêrma exibia, agora, lesões generalizadas, atingindo braços, axilas, pernas.

Escamas epidérmicas obtidas de várias dessas lesões, proporcionaram-nos, sempre, o isolamento de *H. valbyensis*, o que nos leva a admitir a existência de patogenicidade para êsse fungo, no caso em aprêço.

Dessa forma, *Hanseniaspora valbyensis* deverá ser levada em consideração, tembém, no estudo das dermatomicoses.

SUMMARY

This paper deals with the study of two strains of *Hanseniaspora valbyensis* KLOECKER respectively isolated from human cecal appendix contents and from epidermal lesions in one case of dermatomycosis.

A NEW ANTIBIOTIC PRODUCING STREPTOMYCES: STR. BELLUS NOV. SP.

by

P. Margalith & G. Beretta

Research Laboratories Lepetit S.p.A. — Milano

(15.IX.1959)

In many laboratories where screening of streptomycetes for new antibiotics is carried out, little attention is given to strains or species of streptomycetes unless their antibiotic principle can be extracted and the therapeutic value determined. Many of such streptomycetes, however, are found again and again in further screening studies and thus, lead to a considerable amount of rather fruitless work. As has already been pointed out before (1) it would be important if such streptomycetes were adequately described so that when similar strains should turn up those could be immediately identified and their antibiotic potential predicted.

We would like to report, herewith, the finding of a new streptomyces with outstanding morphological characters which make its identity with any previously described microorganism unlikely.

Experimental

The streptomyces was isolated from a soil sample from Lagomaggio (Rimini) and received our collection number A/870. The following description is based on the cultivation of the streptomyces on various nutrient media, unless otherwise stated, according to Waksman (2). The test for the utilization of carbohydrates was made according to Pridham & Gottlieb (3). Color determinations were performed according to Maerz & Paul (4).

Oat-meal agar:	Growth abundant with smooth surface, colorless intense bluish green (velvety) aerial mycelium. Pink soluble pigment, heavy sporulation.
Yeast-extract agar:	Growth abundant with slightly folded surface. Brown vegetative mycelium

	with brown reverse. Whitish aerial mycelium and dark brown pigment.
Emerson-glucose agar:	Heavy growth, rough surface. Pinkish vegetative mycelium, deep pink to pinkish-brown reverse. Aerial mycelium pinkish white with bluish-green spores. Intense reddish brown soluble pigment (5/I/10).
Oat-meal Mix agar 1):	Abundant growth with smooth surface; pinkish brown vegetative mycelium and brownish pink reverse. White aerial mycelium and abundant light blue spores (29/F/1). Pinkish brown soluble pigment (8/H/9).
Bennett's agar:	Growth abundant, pink, with smooth surface. Brown pink reverse. Pinkish white aerial mycelium with abundant bluish green spores. Some light pink soluble pigment with slightly brown tinge. (see fig. 1).
Penassay agar (Difco):	Good growth with smooth surface. Brown vegetative mycelium and brown reverse. Brown pigment. Aerial mycelium and spores absent.
Yeast-extract molasse agar 2):	Growth abundant, smooth, brown, with pinkish brown reverse. Velvety aerial mycelium with abundant greenish blue spores. Brown soluble pigment with pinkish tinge.
Czapek-Dox sucrose agar:	Abundant growth, smooth of light cherry pink (2/I/10 to 5/K/9) colour. Reverse deep cherry pink. Pinkish white aerial mycelium with little sporulation. Light pink soluble pigment (2.J.9) (see fig. 2).

---

1) for composition of media see (5).
2) For composition of medium see (5).

Potato-agar:	Abundant growth, smooth, colorless. Reverse is hyaline to slightly pinkish. Velvety aerial mycelium with good bluish-green sporulation. Traces of pink soluble pigment.
Potato-wedge:	Abundant vegetative mycelium rough colorless to brown. Aerial mycelium and spores absent. Darkening of substrate around growth.
Glucose-asparagine agar:	Good growth with smooth surface, colorless to light pink. Reverse light pink with orange tinge. White aerial mycelium with small amount of bluish spores. Light pink, soluble pigment, with orange tinge.
Glycerol-asparagine:	Good growth, smooth, pinkish, with orange tinge, vegetative mycelium and reverse. Whitish aerial mycelium with some light blue sporulation. Pink to orange soluble pigment (2/C/4).
Nutrient-agar:	Moderate to good growth, hyaline with brownish reverse. Aerial mycelium slight, or absent. Brown soluble pigment.
Pridham's agar:	Moderate growth, colorless, somewhat rough surface and hyaline reverse. Traces of white aerial mycelium. No soluble pigment.
Starch-agar:	Moderate growth, smooth, hyaline, with colorless to pale pink reverse. Some aerial mycelium with bluish green spores. No soluble pigment. Starch hydrolized.
Dextrose-tryptone-agar:	Abundant growth, smooth, pink with brown reverse. Pinkish white aerial mycelium. No sporulation visible. Pinkish to reddish brown soluble pigment.

HICKEY & TRESNER'S cobalt agar (6):	Abundant growth, smooth with reddish brown to brown reverse with or without violet tinge. Heavy, velvet aerial mycelium and abundant penicillium green spores. Some brownish soluble pigment.
Egg-albumin agar:	Moderate growth, colorless vegetative mycelium and reverse. Some white aerial mycelium. Some bluish green spores. No pigmentation.
Tyrosin agar:	Growth scarce.
Ca-malate agar:	Good growth, pink vegetative mycelium with pinkish violet reverse. Abundant greenish blue sporulation. Pinkish violet soluble pigment. Partial digestion of substrate.
Gelatine:	Slow fluidification, brown pigment.
Litmus milk:	Brownish ring of growth. No change in pH of milk.
Nitrates:	Dark brown pigment. No reduction to nitrites.
Cellulose:	Moderate growth on filter paper disks placed on carbohydrate free mineral base agar.
Temperature:	Good growth at both 28° and 37° C. At 37° C pigment formation is more intense, though sporulation seems somewhat inhibited.
Microscopical aspects:	Long flexuous hyphae with monopodial branching of short open hooks (see fig. 3 some closed spirales present. Spores in long straight chains.

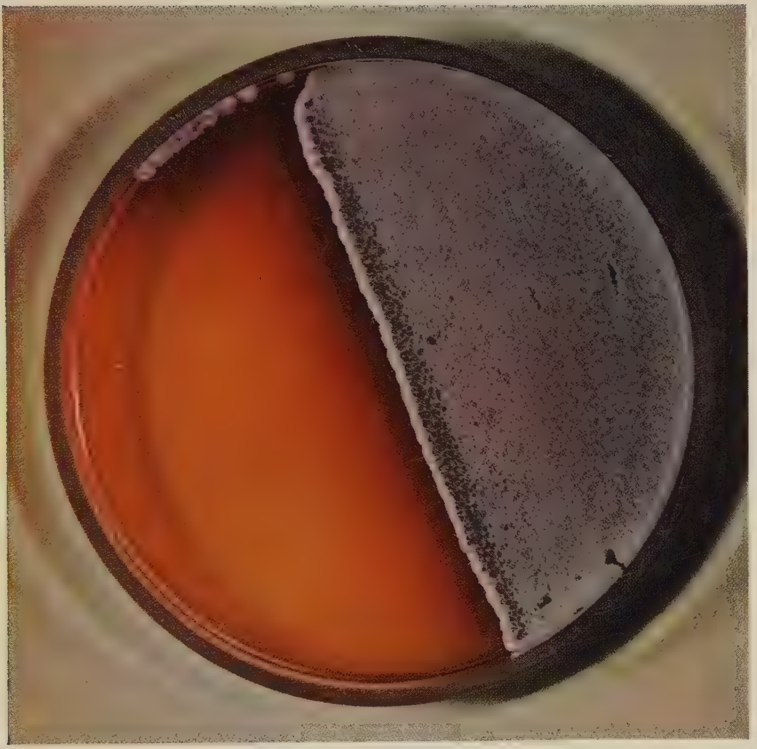


Fig. 1. *Streptomyces bellus* on Bennett's agar medium.

II

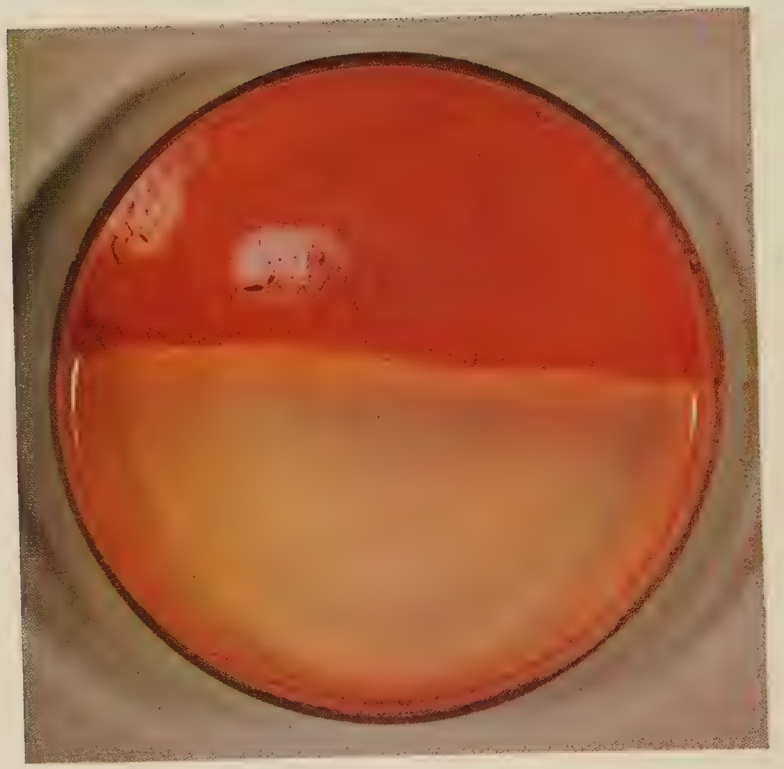


Fig. 2. *Streptomyces bellus* on Czapek-dox agar medium.

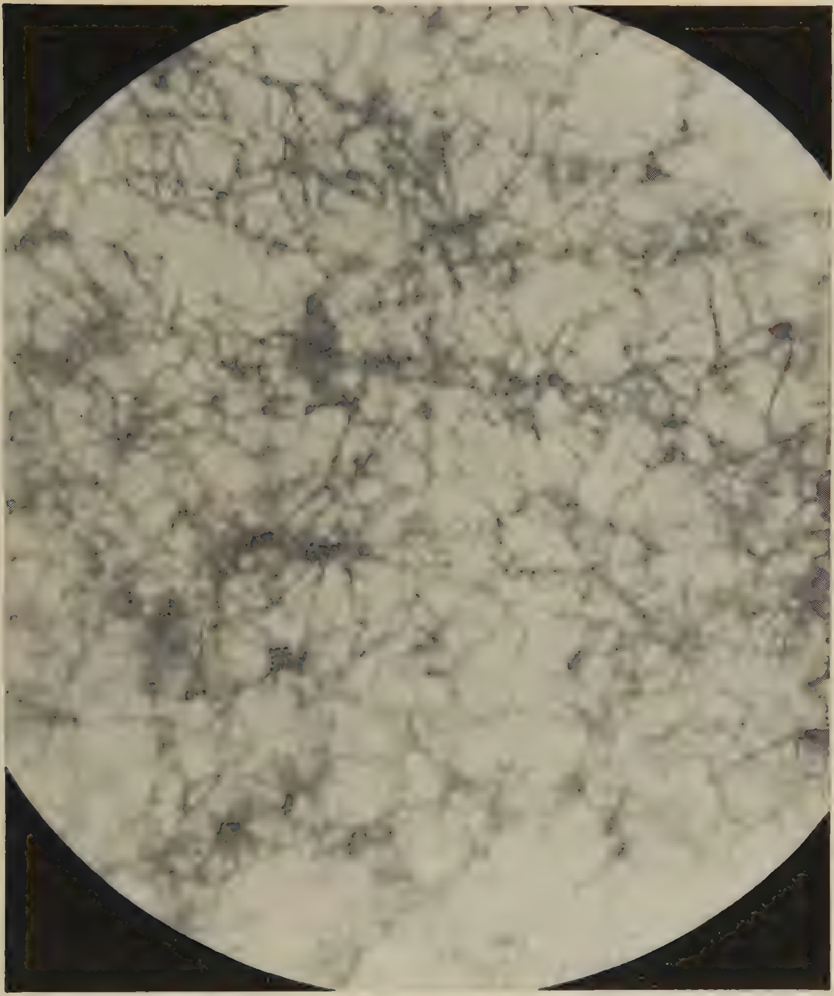


Fig. 3. Aerial mycelium of *Streptomyces bellus* grown on potato agar (× 1000).

For utilization of carbohydrates see Table I.

As seen from the above description, this streptomyces can be easily recognized by the predominating pinkish color of its vegetative mycelium its characteristic soluble pigment as well as the heavy bluish-green sporulation. It should be kept in mind, however, that variation in the intensity of color or abundance of sporulation are rather frequently encountered and cannot be considered as deviations of taxonomic value. Since no similar description of streptomyces could be found in literature (7) we suggest to consider it a new species: *Str. bellus* nov. sp.

TABLE I.

Assimilation of carbon compounds by Streptomyces bellus [1]

Carbon source	Solid media	Liquid media
Lactose	++	+
Galactose	++	+
Arabinose	++	+
Mannose	++	+
Fructose	++	+
Glucose	+	+
Maltose	++	+
Sucrose	++	+
Raffinose	++	+
Rhamnose	++	+
Xylose	—	+
Glycerol	++	+
Mannitol	++	+
Inositol	++	+
Sodium succinate	—	—
Sodium citrate	—	—
Glycine	—	—
Sodium acetate	+	—
Salicin	++	—
Sorbitol	++	—
Inulin	—	±
Dulcitol	—	±

[1] Degrees of growth after five days of incubation are indicated by +, ++. No growth is indicated by —.

TABLE II.

Antimicrobial Spectrum of crude antibiotic A/870 *in vitro*

Test organism	Minimum inhibitory concentration μg/ml
Micrococcus pyogenes var. *aureus*	0,5
Streptococcus faecalis	0,5
Sarcina lutea	0,1
Bacillus subtilis	0,5
Escherichia coli	100
Proteus vulgaris	50
Pseudomonas aeruginosa	100
Klebsiella pneumoniae	50
Candida albicans	100
Trichophyton mentagrophytes	100

FERMENTATION

Conditions for the vegetative inoculum as well as for the final fermentation were similar to those previously described (5). Maximum antibiotic activity was obtained after 48 hrs. of growth. The antibiotic principle was extracted by organic solvents. The antimicrobial activity of the crude product is given in Table II.

Toxicity tests have shown this antibiotic to be devoid of therapeutic value. (DL^{100} mice = 25 mg/kg).

We thank Dr. M. T. TIMBAL and Dr. MAFFII for kindly providing the antibacterial spectrum and toxicity data.

SUMMARY

The description of a new species of Streptomyces, *Str. bellus* nov. sp., is given. The antibiotic principle produced by fermentation procedures is too toxic for therapeutic use.

Bibliography

1. MARGALITH, P. & BERETTA, G. International Symposium for Antibiotics. Prague, May 1959.
2. WAKSMAN, S. A. (1957) The species concept among the actinomycetes with special reference to the genus streptomyces. Bact. Rev. **21,** 1.
3. PRIDHAM, T. G. & GOTTLIEB, D. (1948) The utilization of carbon compounds by some actinomycetales, as an aid for species determination. J. Bact. **56,** 107.
4. MAERZ, A. & PAUL, M. R. (1950) A dictionary of color. 2nd edition McGraw Hill Book Co.
5. MARGALITH, P., BERETTA, G. & TIMBAL, M. T. (1959) Matamycin, a New Antibiotic. I. Biological studies. Antib. and Chem. **9,** 71.
6. HICKEY, R. J. & TRESNER, H. D. (1952) A cobalt containing medium for sporulation of streptomyces species. J. Bact. **64,** 891.
7. Bergey's Manual of Determinative Bacteriology, seventh edition pp. 744—822. The William and Wilkins Company — Baltimore 1957.

TRICHOSPORON FIGUEIRAE N. SP.
ASSINALADO COMO FUNGO INTESTINAL

A. Chaves Batista & Jarbas S. Silveira

(30.VII. 1959)

Instituto de Micologia — Universidade do Recife

Publicação no 171.

O presente trabalho trata do estudo de uma espécie de *Trichosporon* que, por suas características bioquímicas, se distancia das demais espécies dêsse gênero, descritas até então.

A importância dêsse fungo, do ponto de vista da patogenicidade, parẽce achãr-se relacionada com a dinâmica intestinal, provocando manifestações diarréicas.

Dois pacientes portadores dêsse fungo, nas fezes, submetidos a uma dieta restritiva de glucídios e à terapêutica antimicótica, por via oral, com o emprêgo de Mycostãtin, recuperaram a saúde em 20 dias e os retrocultivos de fezes não mais permitiram reisolar êsse *Trichosporon*.

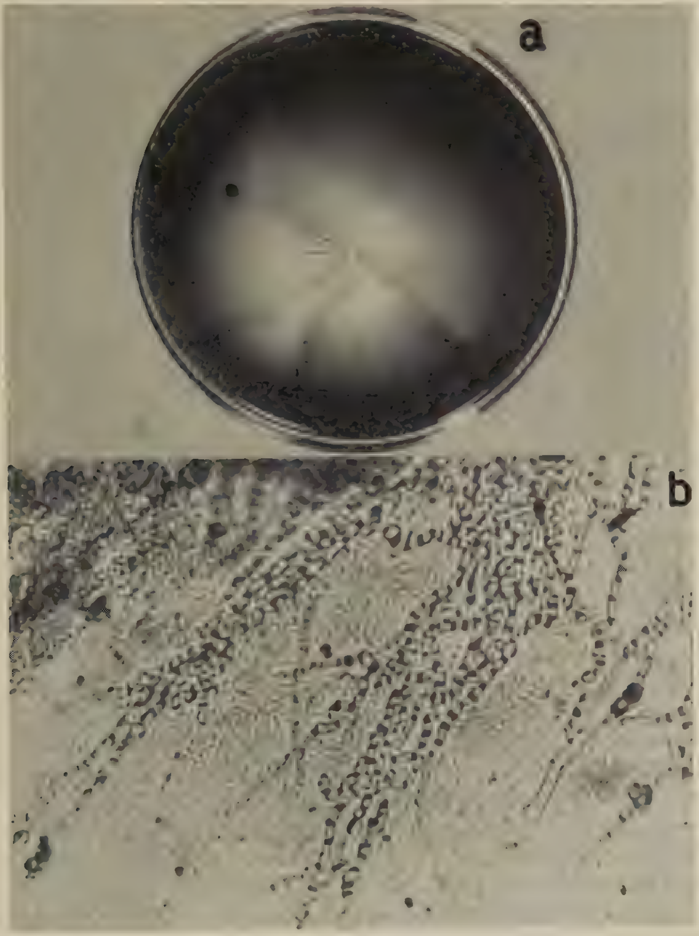
Assim, reconhecemos o organismo em tela como uma nova espécie de fungo leveduriforme, cuja diagnose damos a seguir:

Trichosporon Figueirae Bat. & Silv. n. sp.

(Em homenagem ao Prof. Dr. Antônio Simão dos Santos Figueira, Diretor da Faculdade de Medicina da Universidade do Recife).

Assinalado duas vêzes sôbre fezes humanas de pacientes, que apresentavam distúrbios intestinais, tem as seguintes características:

Cresce bem sôbre sabouraud-agar, de pH = 6,5—6,8, formando colônias circulares, de 5—6 cm de diam., aos 12—15 dias, esbranquiçadas, levemente zonadas, com ligeiros sulcos radiais, filamentosas, parcialmente submersas, opacas, de bordos inteiros, inodoras; reverso creme, de aspecto húmido. Sôbre malte-agar, desenvolvem-se colônias igualmente circulares, 5,5—6,5 cm de diam., aos 12—15 dias e à temperatura ambiente, de aspectio análogo àquelas que se formam sôbre sabouraud agar. Em extrato de malte produz-se película e depósito de sedimento, após 3 dias e à temperatura ambiente; a película é espêssa. Pseudomicélio abundante, micélio ver-



Trichosporon Figueirae BAT. & SILVA. n.sp.
a) Colônia sôbre malte-agar, aos dez dias.
b) Artrosporos.

dadeiro escasso; artrosporos, blastosporos e clamidosporos presentes; artrosporos cilíndricos, 5—8,2 × 3,8—5μ; blastosporos ovóides, elipsóides ou subglobosos, 2,2—5 × 1,6—2,7μ.

Sôbre batata-agar, em lâmina, plantio estriado, há abundante desenvolvimento de micélio artrosporado e blastosporos, além de clamidosporos globosos a subglobosos, intercalares e terminais. O micélio é de hifas hialinas, longas, pouco ramificadas, septadas, predominando a produção de artrosporos catenulados, nas culturas envelhecidas.

Os blastosporos originam-se, frequentemente, na septação das hifas do micélio, aparecendo então isolados, catenulados ou em cachos.

Fermentação — ausente.
Auxonograma — Dextrose (+)
Maltose (+)
Sacarose (—)
Galactose (—)
Lactose (—)

Assimilação de nitrato de potássio: (—)

Etanol — Crescimento satisfatório, com película espêssa e encrespada e pequeno depósito de sedimento.

Leite de litmus — coagulação completa.

Isolado de fezes humanas — (A.J.B. e N.O.B.) do sexo feminino. — Recife — Leg. Dr. ASTIER. Tipo, no. . . . 1028, I.M.U.R

In honorem Prof. Dr. ANTONIO SIMÃO DOS SANTOS FIGUEIRA, Directoris Facult. Medic., Univ. Recifensis.

In sabouraudi-agaro coloniae circulares, albidae, 5—6 cm diam., in 12—15 dies, zonatae, submersae, filamentosae, inodorae, cum reverso cremeo, humido.

In agaro-maltato coloniae 5,5—6,5 cm diam., albidae, similes coloniis in sabouraudi-agaro formantur. In musto maltato pellicula et sedimentum formantur. Pseudomycelium et mycelium; arthrospori cylindrici, 5—8,2 × 3,8—5μ; blastospori ovoidei, ellipsoidei vel subglobosi, 2,2—5 × 1,6—2,7μ; chlamidospori presentes.

In decocto tub. Solani gluc. agaro ("potato-dextrosi") mycelium ex hyphis longis, parum ramosis, septatis, hyalinis compositum. Fermentatio nulla. Dextrosis et maltosis assimilantur. Nitras kalicus non assimilatur. In medio cum alcohole ethylico crescit, cum pellicula et sedimento. Lac litmis precipitat.

In humanis faecibus. (A.J.B. et N.O.B.) Recife — Leg. Dr. ASTIER, 31/3/1958. Typus. 1028, I.M.U.R.

SUMMARY

Trichosporon figueirae BAT. SILV. n. sp. is described as a fungus of intestinal anormal mycoflore.

ISOLATION OF HISTOPLASMA CAPSULATUM FROM AN OIL BIRD (STEATORNIS CARIPENSIS) CAVE IN VENEZUELA

by

Libero Ajello* Tulio Briceño-Maaz** Humberto Campins***
& Jane C. Moore*

(12. XI. 1959)

Discovery of the widespread existence of histoplasmin sensitivity in the population of Williamson County, Tennessee (1—2) inspired an intensive search for *Histoplasma capsulatum* in the soil of that county. During the course of the survey numerous isolates of *H. capsulatum* were obtained (3—5) . A significant correlation was found to exist between the positive soil samples and chicken habitats.

Knowledge of the relationship between areas frequented by chickens and *H. capsulatum* led to the determination that Tingo Maria Fever, a Peruvian pulmonary disease of unknown etiology, was in reality histoplasmosis (6). This fever was known to develop among visitors to "La Cueva de las Lechuzas", a great cavern near the Andean village of Tingo Maria. The cave harbors a large, breeding colony of *Steatornis caripensis*, a chicken-sized nocturnal, fruit eating bird. On the theory that *H. capsulatum* may occur in avian habitats other than those of chickens, soil from the Tingo Maria cave was collected and examined for the presence of *H. capsulatum*. This fungus was recovered from one of four samples (6). Subsequently Arellano & Galvez (7) isolated *H. capsulatum* from two of five students who had developed Tingo Maria Fever after a visit to "La Cueva da las Lechuzas".

Following the Tingo Maria cave findings, a study of *S. caripensis* habitats in other South American countries was initiated to determine whether or not *H. capsulatum* would be present.

The geographic distribution of *S. caripensis* is very limited. This bird has been observed only in British Guiana, Colombia, Ecuador,

---

* Communicable Disease Center, Public Health Service, Department of Health, Education, and Welfare, Atlanta, Georgia.

** Hospital Mene Grande Oil Company, San Tome, Anzoategui, Venezuela.

*** Hospital Central, Barquisimeto, Venezuela.

Peru, Trinidad and Venezuela (8—10). The oil bird was first described by von Humboldt (11) in 1799 from birds collected in the Cave of the Guacharos in the state of Monagas near Caripe, Venezuela. This cave, about 2 miles long, is still inhabited by *S. caripensis*. Arrangements were made to collect soil samples in the Caripe cave in 1956 and 1958.

MATERIALS AND METHODS

The cave is divided into two sections: an anterior section that is readily accessible and an inner portion that is difficult to enter and explore (12). The first section is 3,678 feet long and subdivided into 3 galleries. These are designated as El Salon de los Guacharos (2,685 feet long), El Silencio (622 feet long) and Cuarto Precioso

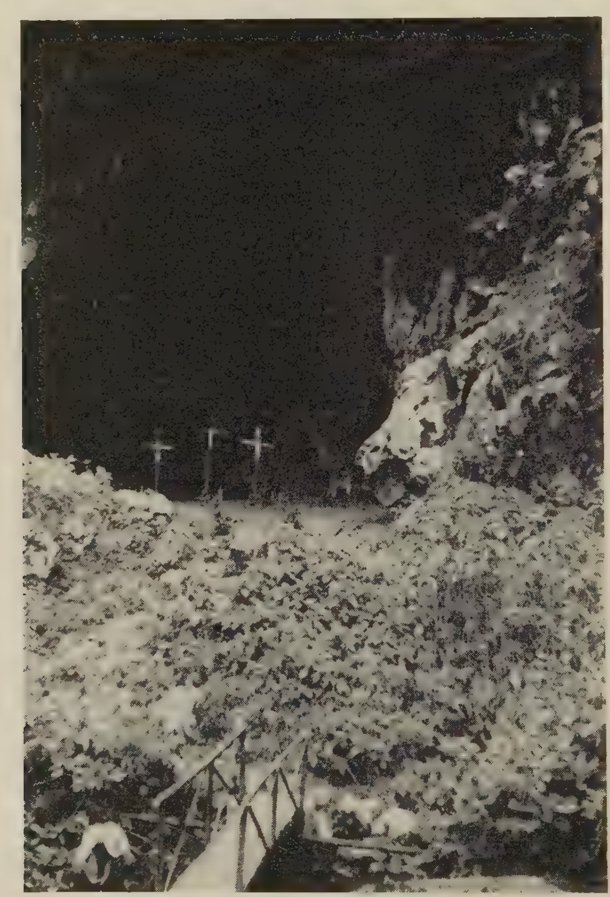


Fig. 1. Entrance to the Guacharo Cave, Caripe, Venezuela.

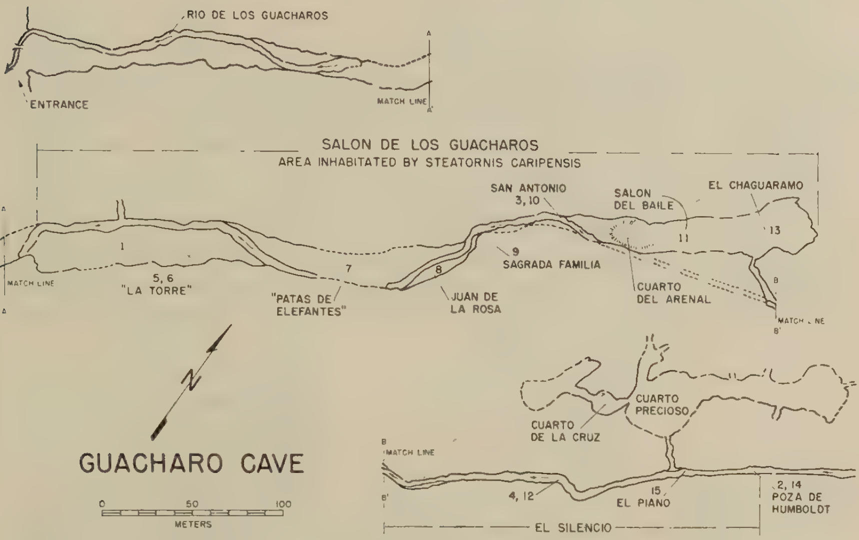

RIO DE LOS GUACHAROS
ENTRANCE
MATCH LINE
A
A'
SALON DE LOS GUACHAROS
AREA INHABITATED BY STEATORNIS CARIPENSIS
SAN ANTONIO
3, 10
SALON DEL BAILE
EL CHAGUARAMO
13
11
9
SAGRADA FAMILIA
CUARTO DEL ARENAL
B
MATCH LINE
B'
A
MATCH LINE
A
1
5, 6
"LA TORRE"
7
"PATAS DE ELEFANTES"
8
JUAN DE LA ROSA
N
GUACHARO CAVE
0
50
100
METERS
CUARTO PRECIOSO
CUARTO DE LA CRUZ
B
MATCH LINE
B'
4, 12
15
EL PIANO
2, 14
POZA DE HUMBOLDT
EL SILENCIO

Fig. 2. Map of the Guacharo Cave.

(370 feet long). The cave's entrance is in the form of a gigantic portico 76 feet high and 85 feet wide (Figures 1—2).

The Guacharo River, a stream whose dimensions vary greatly under the influence of the wet and dry seasons, courses through the cave.

The Salon de los Guacharos has a rather uniform width of 68 feet and a height that ranges from 83 to 99 feet. Massive stalactites but no stalagmites are found within it. One notable cluster of stalactites has been named Patas de Elefantes.

The oil birds nest and roost only in this great gallery. Their territory begins at a point approximately 600 feet from the entrance and extends to, but does not penetrate, El Cuarto del Silencio, 2,319 feet from the entrance. The Cuarto del Silencio is appropriately named, for in that area the cries made by the birds, when disturbed by visitors, cannot be heard. Bats are not found in any part of the cave.

Five soil samples were collected in August, 1956 and 10 in August, 1958 by Dr. T. Briceno-Maaz. The 15 soil collection sites are noted in Figure 1. The specimens were air mailed to the Communicable Disease Center's Mycology Unit in Chamblee, Georgia and processed as soon as received.

There the soils were examined for the presence of systemic human pathogenic fungi by injecting the supernatant from soil suspensions intraperitoneally into mice. Dermatophytes were sought by baiting plates of moistened soil with hair (13).

Mouse Inoculation Procedure:

1. After thorough mixing, 1 heaping teaspoonful of soil was placed in 30 ml of physiological saline that contained 5,000 units of penicillin G and 1,000 units of streptomycin per ml.
2. After vigorous stirring, the soil was allowed to settle for approximately one hour.
3. Five ml of supernatant were then pipetted off and 1 ml injected intraperitoneally into each of 4 mice.
4. Eight weeks after inoculation, the mice were sacrificed and small portions of their liver and spleen were inoculated into tubes of a neutral dextrose agar:

Neopeptone (Difco	1.0 g
Dextrose	1.0 g
Chloramphenicol	0.005 mg
Agar	2.0 g
Distilled water q.s.	100.0 ml

(Adjusted to pH 7 before autoclaving at 120° C for 10 minutes.)

Tissue from each mouse was inoculated into 4 tubes of this medium, 2 tubes with liver and 2 tubes with spleen tissue. A total of 16 tubes therefore were inoculated for each soil sample.

5. The tubes were incubated at 25° C and examined periodically for the development of pathogenic fungi.

Hair-Bait Procedure:

Sterile Petri dishes were half-filled with the soil specimens, and moistened with 15 to 30 ml of sterile distilled water (the amount of water added was dependent upon the nature of the soil sample). Short tufts of autoclaved human hair then were placed upon the surface of the damp soil. The closed Petri dishes were incubated at room temperature in a dark cupboard and examined frequently for the growth of mycelium on the hair filaments. Some of the hairs, overgrown with mycelium, were mounted in lacto-phenol cotton blue and examined under the microscope. Others were cultured on a selective isolation medium that inhibited the growth of saprophytic fungi and bacteria (14).

RESULTS

The first 5 soil samples were negative for systemic pathogenic fungi.* However *Microsporum gypseum* was recovered from a pool of these 5 samples.

From the remaining ten specimens, *H. capsulatum* was isolated from samples 8 and 11, *Allescheria boydii* from soils 6 and 7 and *M. gypseum* from specimens 10, 11 and 13 (Table I).

---

1) These soils were also studied by Dr. H. CAMPINS and found to be negative.

Table I.

Pathogenic fungi isolated from soil collected in the Guacharo Cave, Caripe, Venezuela.

Soil	Collection Site	ISOLATES		
		Allescheria boydii	*Histoplasma capsulatum*	*Microsporum gypseum*
1	Salon de los Guacharos			
2	Poza de Humboldt			Soils 1—5 pooled: +
3	San Antonio			
4	El Silencio			
5	La Torre			
6	La Torre	+		
7	Patas de Elefante	+		
8	Juan de la Rosa		+	
9	Sagrada Familia			
10	San Antonio			+
11	Salon de Baile		+	+
12	El Silencio			
13	El Chaguaramo			+
14	Poza de Humboldt			
15	El Piano			

DISCUSSION

The association of *H. capsulatum* with oil bird habitats, first discovered in Peru (6), confirms establishes the ecological relationship of this fungus to another avian species. *H. capsulatum* now has been isolated from *Steatornis caripensis* habitats as well as those of pigeons, chickens (15) and grackles (16).

This intimate relationship presents a serious public health problem. Numerous outbreaks of acute histoplasmosis have occurred among individuals engaged in cleaning chicken coops, screening and handling chicken or grackle manure or demolishing structures harboring pigeon droppings (17—18).

H. capsulatum occurs in the soil as a saprophyte. There it produces the infectious tuberculate macroconidia that are characteristic of the species (19—20). The fungus is not found in the faeces of birds as they apparently are immune to infections by *H. capsulatum* (4). But it would appear that soil contaminated with bird droppings favors the growth of *H. capsulatum* and enables it to successfully compete with other soil microorganisms.

Erroneous deductions have been drawn from the association of *H. capsulatum* with avian habitats. Menges & Haberman (21) challenged a group of chickens with large inocula of a mixture of mycelium and spores of *H. capsulatum*. The fungus was not recovered from any of the birds. Acquisition of a positive skin test reaction to histoplasmin and the development of positive indirect complement-fixation titres were interpreted to signify infection.

However these reactions could just as well be attributed to sensitization.

The intravenous infection of pigeons and chickens with *H. capsulatum* as described by SCHWARZ and his co-workers (22) involved inoculation with yeast-phase cells rather than macroconidia. It cannot be concluded from this study that domestic fowl are susceptible to infection by *H. capsulatum* under the conditions that prevail in nature.

Domestic fowl do not excrete spores of *H. capsulatum* in their droppings. They do not seed soil with fungus as has been stated editorially in the Journal of the American Medical Association (23). And there is no basis to support the theory discussed by FURCOLOW that *H. capsulatum* is carried by birds (24).

The association of *H. capsulatum* with avian habitats is not absolute for this fungus has been recovered from soils not obviously contaminated with bird droppings. In addition bat habitats have yielded *H. capsulatum* (15).

Caves in several parts of the world have proven to be dangerous to speleologists and other visitors due to the precense of *H. capsulatum* in the cave soil. Such episodes have been reported in East Africa (25), Peru (6), South Africa (26), United States (27-28) and Venezuela (29-32). The histoplasmosis hazard is so well recognized in Peru that a notice has been posted at the entrance of the Tingo Maria Cave warning visitors of the danger of contracting histoplasmosis if the cave is entered (33).

The Venezuelan Society of Natural Sciences has published 2 communications by Dr. EUGENIO DE BELLARD PIETRI pointing out the danger of contacting histoplasmosis in certain caves (30, 34).

A list of caves and their status in relation to *H. capsulatum* was prepared (34). Seventeen were classified as free of *H. capsulatum*, including the Caripe Guacharo Cave and 6 were cited as harboring *H. capsulatum*. This classification is not completely based on soil studies and thus is subject to error. This present study, in fact, reveals that the Caripe cave does harbor *H. capsulatum* and presents a potential hazard to visitors. But it is important to note that up to now no cases of histoplasmosis have been traced to the Guacharo Cave despite the fact that it has been visited by many people through the years. The absence of infections may be due to the absence of dust in the cave. Humidity in the cave is high since water constantly drips from the roof and walls and the Guacharo river, which flows through the cave, never dries even in the dry season. The floor of the Salon de los Guacharos is composed of a mixture of bird guano and mud.

From the public point of view, caves with bat and oil bird populations are potential sources of *H. capsulatum* infections. Public Health Departments and speleological societies should warn all persons that explore caves of the inherent danger of contracting histoplasmosis.

SUMMARY

1. *Histoplasma capsulatum* was recovered from two of fifteen soil specimens collected in a Venezuelan cave. This cave, located near Caripe in the state of Monagas, harbors a large colony of the oil bird, *Steatornis caripensis.*
2. This finding confirms the previous discovery in Tingo Maria, Peru, of the association of *H. capsulatum* with oil birds.
3. The relationship of *H. capsulatum* with animal habitats is discussed along with the public health importance of this association.

ZUSAMMENFASSUNG

1. *Histoplasma capsulatum* ist in zwei Bodenproben aus fünfzehn, die einer Höhle in Venezuela entnommen wurden, gefunden worden. Diese Höhle, die sich in der Nähe von Caripe, im Staate Monagas befindet, herbergt eine grosse Kolonie von ,,Fettvögeln'', *Steatornis caripensis.*
2. Dieser Befund bestätigt die Vergesellschaftung von *H. capsulatum* mit den ,,Fettvögeln'', wie es vorher bereits in Tingo Maria, Peru, entdeckt worden ist.
3. Die Beziehung von *H. capsulatum* zum tierischen Habitat wird diskutiert unter Betonung der Wichtigkeit dieser Vergesellschaftung vom Standpunkte des öffentlichen Gesundheitswesens.

REFERENCES

1. Christie, A. & Peterson, J. C. (1945), Pulmonary calcification in negative reactors to tuberculin. Amer. J. Pub. Health **35:** 1131—1147.
2. Zeidberg, L. D., Dillon, A. & Gass, R. S. (1951), Some factors in the epidemiology of histoplasmin sensitivity in Williamson County, Tennessee. Amer. J. Pub. Health **41:** 80—89.
3. Ajello, L. & Zeidberg, L. D. (1951), Isolation of Histoplasma capsulatum and Allescheria boydii from soil. Science **113:** 662—663.
4. Zeidberg, L. D., Ajello, L., Dillom, A. & Runyon, L. C. (1952), Isolation of Histoplasma capsulatum from soil. Amer. J. Pub. Health **42:** 930—935.
5. Zeidberg, L. D. & Ajello, L. (1954), Environmental factors influencing the occurrence of Histoplasma capsulatum and Microsporum gypseum in soil. J. Bact. **68:** 156—159.
6. Lazarus, A. S. & Ajello, L. (1955), Aislamiento de Histoplasma capsulatum del suelo de una cueva en el Perú. Rev. Med. Exp. Lima **9:** 5—15.
7. Arellano, R. C. Z. & Galvez, J. B. (1955), El agente etiológico de la fiebre de ,,Tingo Maria''. An. Fac. Med. Lima **38:** 1092—1098.
8. Hollister, G. E. (1926), The Guacharo or oil bird of the Arima Gorge. Zool. Soc. Bull. **29:** 139—145; 160—161.
9. Griffin, D. R. (1953), Acoustic orientation in the oil bird, Steatornis. Proc. Nat. Acad. Sci. **39:** 884—893.
10. De Bellard, E. P. (1957), El Guacharo. Bol. Soc. Venezolana Ciencias Nat. **18:** 3—41.
11. Humboldt, A. de & Bonpland, A. (1814), Voyage de Humboldt et Bonpland. Première Partie. Relation Historique. Volume 1, Chapter 7: Couvent de Caripe-Caverne du Guacharo-Oiseaux nocturnes. 409—431. Chez G. Dufour et Comp., Paris.
12. De Bellard, E. P. (1952), The Guacharo Cave. The American Caver **14:** 15—18.

13. Vanbreuseghem, R. (1952), Technique biologique pour l'isolement des dermatophytes du sol. Ann. Soc. Belge Med. Trop. **32:** 173—178.
14. Georg, L. K., Ajello, L. & Papageorge, C. (1954), Use of cycloheximide in the selective isolation of fungi pathogenic to man. J. Lab. Clin. Med. **44:** 422—428.
15. Emmons, C. W. (1958), Association of bats with histoplasmosis. Pub. Health Rep. **73:** 590—595.
16. Ajello, L. Unpublished data.
17. Ajello, L. (1958), Geographic distribution of Histoplasma capsulatum. Mykosen **1:** 147—155.
18. Hosty, T., Ajello, L., Wallace, G. D., Howell, J. & Moore, J. (1958), A small outbreak of histoplasmosis. Amer. Rev. Tb.Pul. Dis. **78:** 576—582.
19. Emmons, C. W. (1949), Isolation of Histoplasma capsulatum from soil. Pub. Health Rep. **64:** 892—896.
20. Ajello, L. (1954), Occurrence of Histoplasma capsulatum and other human pathogenic molds in Panamanian soil. Amer. J. Trop. Med. Hyg. **3:** 897—904.
21. Menges, R. W. & Habermann, R. T. (1955), Experimental avian histoplasmosis. Amer. J. Vet. Res. **16:** 314—320.
22. Schwarz, J., Baum, G. L., Wang, C. J. K., Bingham, E. L. & Rubel, H. (1957) Successful infection of pigeons and chickens with Histoplasma capsulatum. Mycopath. et Mycol. Appl. **8:** 189—193.
23. Editorial (1958), Histoplasmosis. J. Amer. Med. Ass. **186:** 2272.
24. Furcolow, M. L. (1958), Recent studies on the epidemiology of histoplasmosis. Ann. N.Y. Acad. Sci. **72:** 127—164.
25. Manson-Bahr, P. E. C. (1958), Histoplasmosis in East Africa. E. Afr. Med. J. **35:** 612—630.
26. Murray, J. P., Lurie, H. I., Kaye, J., Komins, C., Borok, R., & Way, M. (1957), Benign histoplasmosis (Cave Disease) in South Africa. S. Afr. Med. J. **31:** 245—253.
27. Grayston, J. T. & Furcolow. M. L. (1953), The occurrence of histoplasmosis in epidemics: Epidemiological studies. Amer. J. Pub. Health **43:** 665—676.
28. Furcolow, M. L. & Grayston, J. T. (1953), Occurrence of histoplasmosis in epidemics: etiologic studies. Amer. Rev. Tuberc. **68:** 307—320.
29. Campins, H., Zubillago, C., Gomez, L. & Dorante, M. (1955), Estudio de una epidemia de Histoplasmosis en el Estado Lara, Venezuela, Cac. Med. Caracas **57:** 85—109.
30. De Bellard, E. P. (1955), Grave enfermedad identificada en varias cuevas de Venezuela. Bol. Soc. Ven. Ciencias Nat. **16:** 209—211.
31. De Bellard, E. P. Briceno, L. I., Pardo, I., Requena, A. & Pollak, L. (1956), La histoplasmosis en Venezuela. Cac. Med. Caracas nos. **3—5:** 191—. 208.
32. De Montemayor, L., Heredia, B. O., & De Bellard, E. P. (1958), Aislamiento del Histoplasma capsulatum en el suelo de dos cavernas en Venezuela. Nuevas técnicas de investigación por „metodo de flotacion". Rev. Sanidad y Aist. Social **23:** 39—54.
33. Lazarus, A. S. Personal communication.
34. De Bellard, E. P. (1957), La histoplasmosis y los cuevas de Venezuela. Bol. Soc. Ven. Ciencias Nat. **18:** 181—185.

CANDIDA IN VITRO AND IN VIVO

1. CLINICAL ASPECTS:

by

C. Howard Whittle[1])

2. EXPERIMENTAL ASPECTS:

by

G. Austin Gresham[2])

(24.XI.1959)

1. Clinical Aspects

Candida cultivated in routine sugary media grows like a yeast with smooth moist white or creamy glistening colonies with a yeasty smell. Culture mounts show that the organism is multiplying by simple budding with formation of blastospores with little or no hyphal growth. Yet in clinical medicine *Candida* is seen chiefly in mycelial form, suggesting that this is the form best adapted to survival in human tissues.

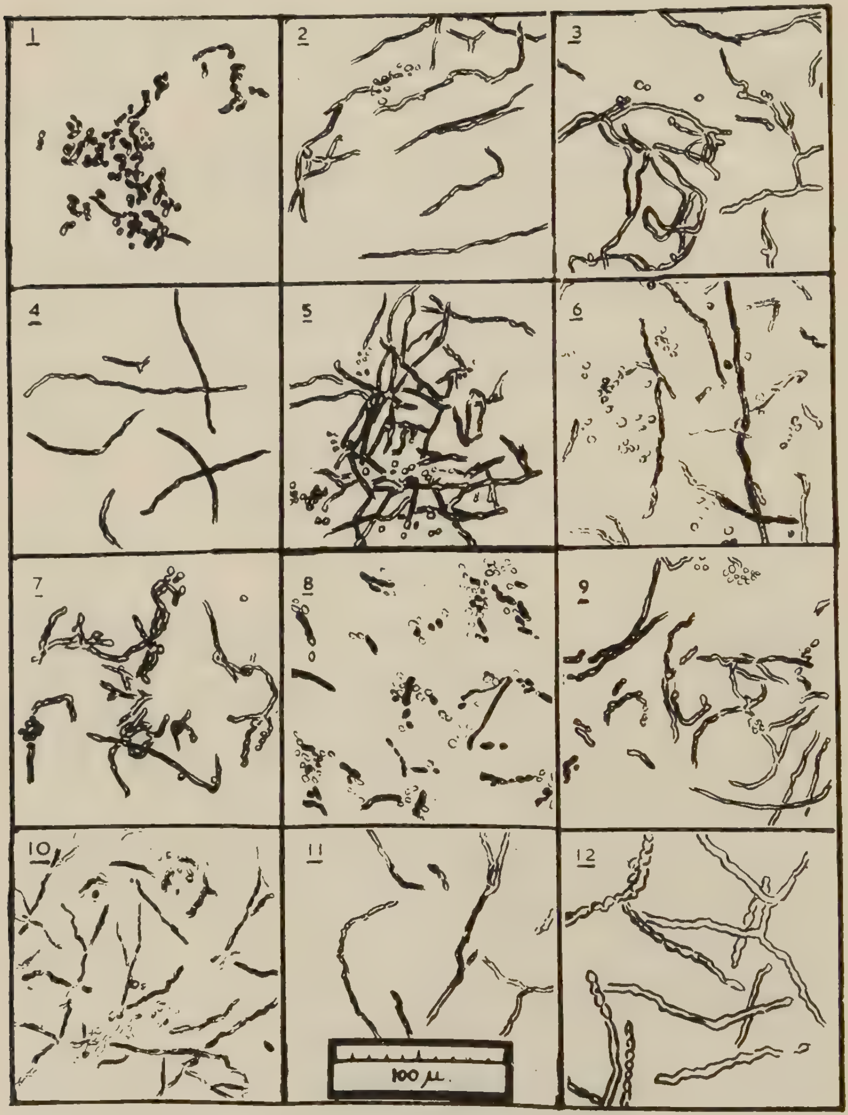
In spite of this, and for various reasons, the idea has been expressed or assumed that *Candida* grows in the tissues in yeast form, perhaps because such fungi as *Histoplasma* and *Cryptococcus* invade tissues in the blastospore or yeast phase.

Dimorphism in the *Candida* genus has been the subject of much study in the past, (Wickerham & Rettger, 1939; Henrici, 1940; Skinner, 1947; Scherr & Weaver, 1953) but even now there is disagreement as to the phase in which it invades. Scherr & Weaver, Winner (1958), and others have followed Mackinnon's lead (Mackinnon, 1940) in regarding the yeast phase as the invasive form, but this was largely because of his discovery of a strain of *Candida albicans*, originally pathogenic, which had become non-pathogenic in its mycelial form, a form which proved to be a stable mutant which could only temporarily be reversed to the yeast phase. Nevertheless we find it difficult to accept such a view in

---

[1]) Consultant Dermatologist, United Cambridge Hospitals and Associate Lecturer in Dermatology, University of Cambridge, England.

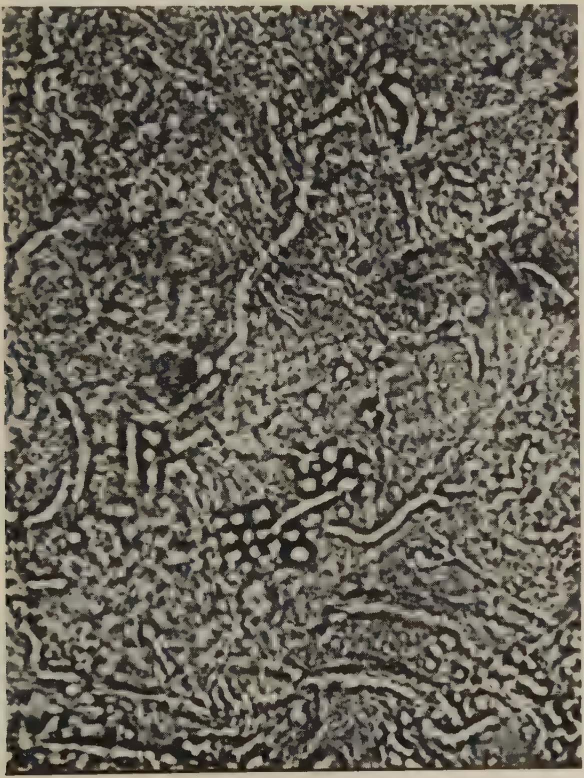
[2]) University Lecturer, Department of Pathology, University of Cambridge, England.



1. *Candida albicans*: perionychia — L. thumb fold.
2. *Candida albicans*: perionychia — L. index nail fold. (16 colonies).
3. *Candida albicans*: perionychia — L. index nail fold. (100 colonies).
4. *Candida albicans*: perionychia — R. ring nail fold.
5. *Candida albicans*: angular stomatitis. (27 colonies pure).
6. *Candida albicans*: moniliasis of breast areola. (15 colonies).
7. *Candida albicans*: intertrigo groin. (1 colony).
8. *Candida tropicalis*: perionychia — R. index nail fold. (16 colonies).
9. *Candida albicans*: intertrigo toe cleft (4th) (Moderate growth).
10. *T. interdigitale*: intertrigo toe cleft.
11. *T. rubrum*: intertrigo toe cleft.
12. *E. floccosum*: intertrigo toe cleft.

face of our fairly constant routine clinical findings: and we are supported in our belief that *Candida* is mycelial in its invasive phase by such observers as REDAELLI (1924), WICKERHAM & RETTGER, FOSTER (1947), AINSWORTH (1955), ROGERS (1957), KAPICA & BLANK (1957), WALKER (1958), YOUNG (1958), (e.g. "The mycelial rather than the yeast phase appears to be the infective form of the pathogen" - YOUNG).

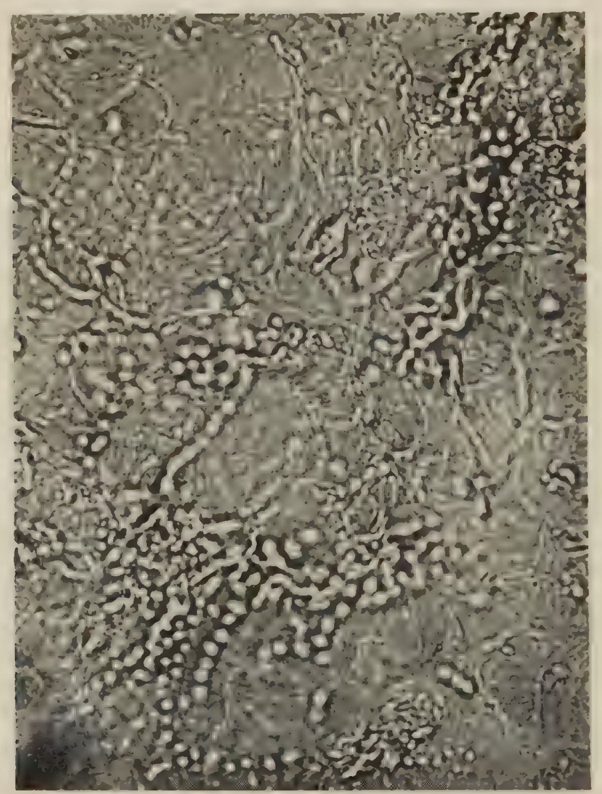
This mycelial form is well illustrated in the accompanying camera lucida drawings and photomicrographs of routine scrapings mounted in 30% potassium hydroxide and taken from lesions in which *Candida* was the sole or the predominating micro-organism. (Figures 1—8). I have no proof that these strains were pathogenic, but they were present fairly constantly and often in abundance, and came from such lesions as have for many years been attributed to *Candida* infection. Most of the material has been obtained from inflamed nail-folds, perionychia, (WHITTLE, MOFFATT & DAVIS, 1959, WHITTLE & GRESHAM 1959), but scrapings from angular stomatitis,



13. *C. albicans* in scales — photomicrograph (× 450).

thrush, moniliasis of the napkin area in infants, intertrigo of the submammary folds, groins, internatal cleft, and toe-clefts gave the same picture (Figures 1—8): and the cultures yielded *Candida albicans* in amount varying from one to one hundred colonies on slope culture with no other fungi appearing.

The resemblance between *Candida* and the dermatophytes in vivo is well illustrated by the finding of mycelium in scrapings from the toe-clefts of four different patients: the first grew *Candida albicans* (Figure 9), the second *T. interdigitale* (Figure 10), the third *T. rubrum* (Figure 11), and the fourth *E. floccosum* (Figure 12). In vivo they may look much alike. Figures 13 and 14, photomicrographs, exemplify the similarities between *C. albicans* and *T. interdigitale* in scales.



14. *T. interdigitale* in scales — photomicrograph (× 450).

Discussion

C. albicans and *C. parapsilosis* can attack keratin in vitro, using it as sole source of nitrogen (Kapica & Blank, 1957, 1958). *C.*

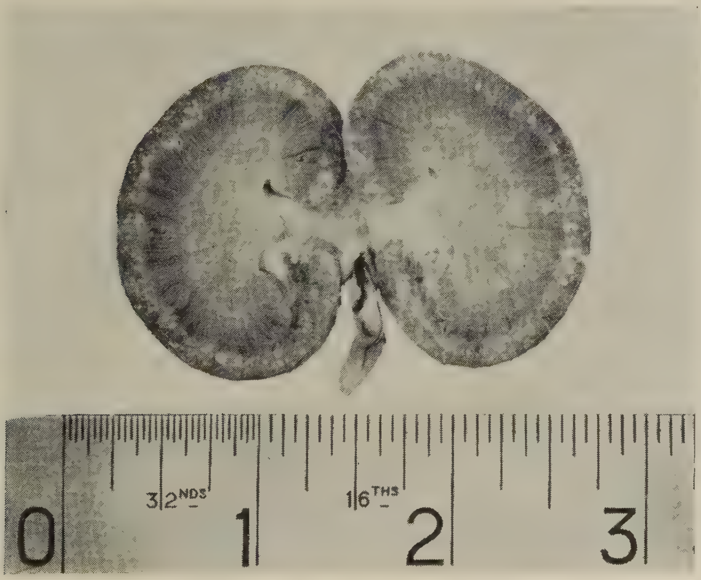
albicans in the process forms abundant mycelium after about 30 days. We believe it produces true hyphae by septation of the filament, (WICKERHAM & RETTGER), which is capable of transporting food from one part of the plant to another. It is thus closely linked phylogenetically to the moulds, in which sugar fermentation is a sign of disordered metabolism, (FOSTER). *Candida* in vivo behaves like the dermatophytes which are essentially mycelial in their invasive form.

ACKNOWLEDGMENTS

I wish to thank Dr. JACQUELINE WALKER of the London School of Hygiene and Tropical Medicine for her help in identifying all the cultures; also Messrs. S.W. PATMAN and S.W. VINCE for the photomicrographs and the camera lucida photographs respectively.

2. EXPERIMENTAL ASPECTS

Yeast is a word originally used to describe the process of alcoholic fermentation (Dutch, gist = foam; German, Geist). SCHWANN and CAGNARD-LATOUR found budding cells in alcoholic fermentation sediments and called these yeasts; they are ascosporogenous. Later morphologically similar, but asporogenous organisms such as *Candida* were also called yeasts. So it was that we have come to regard

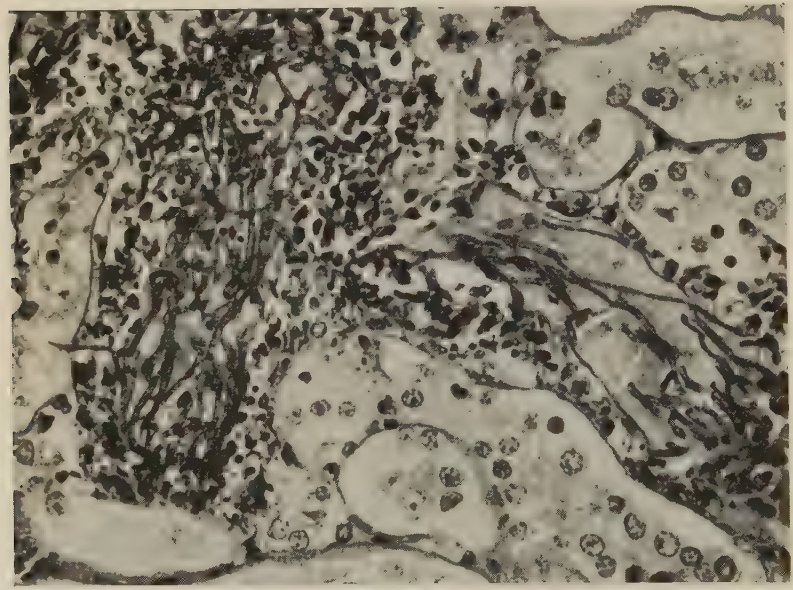


15. Experimental lesions in renal cortex of rabbit.

the yeast form of *Candida* as being its "natural" form. It is the saprophytic phase of the organism. I shall present some experimental evidence that the parasitic phase of *Candida albicans* and of *C. tropicalis* is mycelial and furthermore the assumption of a hyphal form is essential for pathogenicity.

Candida albicans is usually a surface saprophyte and an occasional parasite in man. A study of the evolution of thrush lesions in children (ROGERS, 1957) has shown that the appearance of clinically detectable lesions is associated with the finding of hyphae in the mucous membrane. The significance of such hyphae is better appreciated by a study of experimental infections with *Candida* species in a variety of animals.

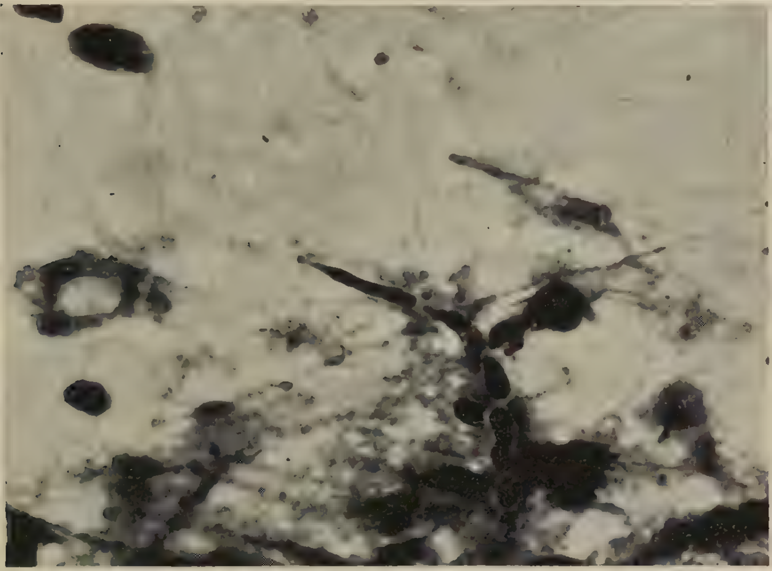
Intravenous inoculation of *Candida albicans* will kill the rabbit; at necropsy the kidneys are studded with white dots confined, at first, to the cortex, (Figure 15).



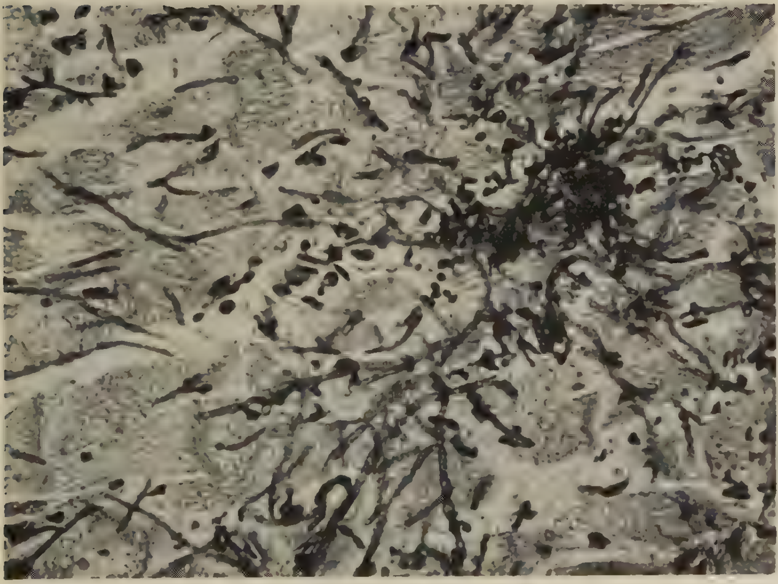
16. Hyphal growth extending into Bowman's capsule and into proximal convoluted tubules.

A study of the evolution of the lesions shows first that the organism is present as yeast forms in cortical capillaries.

If the animal be killed on the third or fourth day hyphae can be seen in histological preparations extending into Bowman's capsule and into the proximal convoluted tubules (Figure 16). Impression preparations at this time show numerous hyphae and the presence of a capsule, (Figure 17).



17. Hyphae with capsule from renal lesion.



18. Invasion of tissue by penetrating hyphae.

Cortical capillaries are packed with blastospores and hyphae and also contain thrombi. A histological examination of other organs at this time shows foci of blastospores in capillaries in heart, brain and many other organs. From these foci invasion is accomplished by penetrating hyphae, (Figure 18).

If a rabbit be allowed to die "naturally" rather than be killed after an intravenous inoculation of *C. albicans* the blood urea will rise up to 300 or 400 mg/100 ml and the urine output will fall. The plasma clearance of P.A.H. drops to very low levels. It appears that this is an obstructive oliguria.

Here then is the evidence which favours the view that pathogenicity of *C. albicans*, in the rabbit, depends on mycelial formation:

(i) Drop in Renal blood flow, oliguria and raised blood urea:
(ii) Histological finding of mycelial masses in vessels:
(iii) Formalin killed *Candida albicans* or *C. guilliermondi* or *C. parapsilosis* will not kill intravenously:
(iv) Simkin's experiments with glass spheres (50—180μ) which, though larger than the glomerular capillaries, could be recovered from the renal vein after injection into the renal artery. These experiments suggest that mechanical blocking by blastospores cannot entirely explain the renal changes.

Vascular obstruction may be due to mycelial mats; thrombosis may be brought about by the capsular aggressin. Purely mechanical obstruction of vessels is, of course, a usual phenomenon in plant pathology. The "wilts" are brought about in this way.

Candida tropicalis, parapsilosis and *guilliermondi* did not kill the rabbit. Post mortem examination of killed animals showed blastospores in glomerular capillaries and in vessels in other organs as revealed by organ culture. *C. tropicalis* will however kill the rat and mouse. Here again mycelium and a capsule are found.

This brief account illustrates an attempt to probe into that all-too-confused field of microbial virulence, as distinct from host resistance. Two factors seem to be of importance:

(i) Ability to form mycelium:
(ii) Capsular aggressins.

Persistent M. variants of *Candida* are said to be non-pathogenic and this is a provoking thought for further research. The appearance of granulomatous hypersensitivity reactions to *Candida* is another stimulus to investigation particularly of the antigenic structure of the organism.

References

AINSWORTH, G. C. (1955) "Pathogenicity of Fungi in Man and Animals" Page 242 in "Mechanisms of Microbial Pathogenicity". edited by J. W. Howie & A. J. O'Hea. Cambridge University Press.

FOSTER J. W. (1947) Introspection on Moulds. Bact. Rev. **11,** 167.

HENRICI, A. T. (1940) Characteristics of Fungous Diseases. J. Bact. **39,** 113.

KAPICA, L. & BLANK, F. (1957) Growth of Candida albicans on keratin as sole source of nitrogen. Dermatologica. **115,** 81.

KAPICA, L. & BLANK, F. (1958) Growth of Candida parapsilosis on keratin as sole source of nitrogen. Dermatologica. **117,** 433.

MACKINNON, J. E. (1940) Dissociation in Candida albicans. J. infect. Dis. **66,** 59.

REDAELLI, P. (1924) Experimental moniliasis. J. Trop. Med. & Hygiene. **27,** 211.

ROGERS, K. B. (1957) Candida in thrush. J. Clin. Path. **10,** 406.

SCHERR, G. H., & WEAVER, R. H. (1953) The Dimorphism phenomenon in yeasts. Bact. Rev. **17,** 51.

SKINNER, C. E. (1947) The Yeast-like Fungi: Candida & Brettanomyces. Bact. Rev. **11,** 227.

WALKER, J. (1958) Personal communication.

WHITTLE, C. H., MOFFATT, J. L. & DAVIS, R. A., (1959) Paronychia or perionychia: aetiological aspects. Brit. J. Derm. **71,** 1.

WHITTLE, C. H. & GRESHAM, G. A. (1960) Candida in keratin of nail folds. Proc. of Symposium on Progress in Biological Sciences in Relation to Dermatology: Cambridge University Press: (in the press). p. 168.

WICKERHAM, L. J. & RETTGER, L. F. (1939) A taxonomic study of Monilia Albicans with special emphasis on morphology and morphological variation. J. Hyg. & Trop. Med. **42,** 174—177, 187—192, 204—216.

WINNER, H. I. (1958) Personal Communication.

YOUNG, G. (1958) The process of invasion and persistence of Candida albicans injected intraperitoneally into mice. J. infect. Dis. **102, 114.**

LE PROBLÈME DE L'EXISTENCE ET DE LA MISE EN ÉVIDENCE DES FERMENTS KÉRATINOLYTIQUES CHEZ LES DERMATOPHYTES

par

RADU EVOLCEANU & MARIA LAZAR

(*Section Dermatologique de l'Institut de Thérapeutique de Bucarest. Directeur: Prof.* ST. G. NICOLAU, *membre de l'Académie Roumaine*)

(30.VIII.1959)

Les dermatophytes sont des champignons parasites dont la kératinophylie est connue depuis longtemps. Leur avidité pour la kératine, bien que non absolue, est tout de même réelle. Elle a servi à VANBREUSEGHEM (11) d'isoler ces champignons des couches superficielles du sol, en mettant des cheveux en contact avec de la terre. Néanmoins, le problème de l'existence, chez les dermatophytes, des ferments à pouvoir kératinolytique, n'a pas encore trouvé une solution satisfaisante. Depuis les travaux de ROBERTS en 1889 (6), de TRUFFI en 1901—1905 (10) et surtout de VON MALLINCKRODT-HAUPT en 1928 (5), l'on sait que les dermatophytes élaborent une protéase analogue à la trypsine, mais pas à la pepsine et, moins encore, une protéase à pouvoir kératinolytique.

D'après ce qu'il paraît, en 1950, STAHL et coll. (8) ont réussi à obtenir une digestion partielle de la kératine par le *Microsporum gypseum*, non seulement d'une façon directe, mais aussi indirectement, à l'aide d'un filtrat des cultures de ce champignon. Cela prouverait l'existence, chez ce dermatophyte, d'une exoenzyme kératinolytique, c'est à dire d'une exoenzyme autre que la trypsine.

Jusqu'à présent, on connaît peu de protéases dont l'action lytique sur les kératines véritables („eukératines") soit bien démontrée. Le règne animal nous en offre un seul exemple, à savoir celui du ferment contenu dans l'intestin de la mite *Tineola bisellida* (7) au stade larvaire. L'action de cette protéase, dont la température optime est 40° C et le pH optime est 9,3, paraît être précédée par l'action d'un réducteur qui transformerait un groupement moléculaire — S — S — en deux groupements — SH. Dans le règne végétal, c'est la papaïne (2), contenue dans les fruits de l'arbre tropical *Carica papaya*, qui exerce une certaine action kératinolytique. Mais, c'est surtout parmi les microorganismes, qu'on trouve des représentants chez lesquels cette faculté de lyser la kératine est le mieux développée. La propriété d'attaquer la kératine a été trouvée chez les Actinomycètes (3) et, dernièrement, KAPICA & BLANK (4) l'ont mise en évidence chez les espèces de *Candida*.

Comme il a été dit plus haut, bien que la kératinophylie des dermatophytes soit bien connue, le mécanisme de la digestion de la kératine par ces champignons est resté assez nébuleux, puisqu'on n'a pas réussi à trouver une méthode appropriée, capable de mettre en évidence, d'une façon constante, leur enzyme kératinolytique. Dans le travail de STAHL et coll., cité au début de l'article, mention en est faite seulement en ce qui concerne le *Microsporum gypseum*. C'est dans le but d'obtenir des dates supplémentaires, que nous avons entrepris nos recherches. Il nous fallait préciser, auparavant, lesquels parmi les dermatophytes sont les plus kératinophyles in vitro. En répétant les expériences de VANBREUSEGHEM sur des cheveux isolés, nous avons trouvé que les espèces qui ont le plus d'avidité pour la kératine sont les espèces dites vivaces, à savoir le *Microsporum gypseum*, le *Microsporum lanosum*, le *Ctenomyces mentagrophytes*, l'*Achorion quinckeanum* et le *Ctenomyces interdigitalis*, les plus actives étant les deux premières. Pour effectuer nos essais, nous devrions donc utiliser surtout ces espèces.

EXPÉRIENCES PERSONNELLES

Afin de préciser si, parmi les protéases élaborées par les dermatophytes, ne s'y trouvent pas des exoenzymes capables de digérer la kératine, nous avons imaginé un modèle expérimental extrêmement simple. Nous avons préparé, tout simplement, des suspensions de kératines finement triturées, que nous avons distribué dans des petits tubes (tubes à hémolyse). Nous y avons ensemencé les dermatophytes suivants: *Microsporum gypseum*, *Microsporum lanosum*, *Trichophyton gypseum* (*Ctenomyces mentagrophytes*), *Trichophyton violaceum*, *Ctenomyces interdigitalis*. A l'intention de porter nos expériences sur des kératines véritables, c'est à dire autant que possible non modifiées, nous n'avons pas utilisé des kératines purifiées mais des organes presque exclusivement formés par de la kératine, à savoir: de cheveux, de la laine, de la corne de boeuf. Ces organes, après avoir été dégraissés par l'acétone, ont été finement triturés. Le titre de nos suspensions a été de 0,02 %. Les tubes ensemencés ont été maintenus à la température de la chambre (18°—20° C). Après 4—5 jours, des petits flocons de culture se sont formés dans les tubes où l'on avait introduit le *Microsporum gypseum*, le *Microsporum lanosum*, le *Ctenomyces mentagrophytes* et le *Ctenomyces interdigitalis*, et la suspension est devenue limpide. On n'y voyait plus des particules de kératine, mais seulement quelques flocons de culture. Il nous paraît difficile d'admettre que toutes ces particules fussent attirées par les sus-dits flocons, d'autant plus que l'épreuve à la ninhydrine, effectuée avec des filtrats de ces cultures, avait été fortement positive. Dans les tubes qui avaient été ensemencés avec le *Trichophyton violaceum*, bien que les flocons de culture se soient développés, la suspension est restée trouble. Mais l'épreuve à la ninhydrine a été positive (moins fortement que dans les cas précédents) ce qui prouve qu'une digestion, au moins partielle, de la kératine, s'est tout de même produite.

Afin d'avoir une preuve sûre de la digestion indirecte de la kératine par les dermatophytes, autrement dit, afin de démontrer l'existence chez ces champignons d'une exoenzyme capable de lyser la kératine, nous avons séparé, dans chacun de nos tubes, la suspension, en deux étages, à l'aide d'un bouchon d'ouate hydrophyle, placé à mi-hauteur de la colonne liquide. On a ensemencé l'étage supérieur, l'expérience portant sur les mêmes espèces de dermatophytes comme ci-dessus. Ce bouchon était suffisament épais (1 cm environ) pour ne pas permettre la contamination de l'étage inférieur, contamination que l'on aurait d'ailleurs immédiatement observé, grâce à l'apparition des flocons de culture à ce niveau. En même temps, le bouchon permettait le libre passage, entre les deux étages, des substances dissoutes. En empêchant la propagation des cultures à l'étage inférieur, ces simples bouchons d'ouate ont remplacé avec succès les filtres poreux habituels qui, dans ce modèle expérimental, auraient nécessité une commande spéciale.

Comme précédemment, on a préparé les suspensions avec des cheveux, de la laine et de la corne de boeuf, finement triturés, ayant le même titre (0,02 %). Les tubes ont été ensemencés et maintenus à la température de 20° C environ (la température de la chambre). Après 15 jours, la suspension contenue dans les tubes est devenue limpide, non seulement à l'étage supérieur, où les particules de kératine avaient été remplacées par des flocons de culture, mais aussi à l'étage inférieur, qui ne présentait aucune trace de mycélium [1]). Ce phénomène ne s'est produit que dans les tubes où l'on avait introduit les dermatophytes „vivaces", à savoir: le *Microsporum gypseum*, le *Microsporum lanosum*, le *Ctenomyces mentagrophytes* et le *Ctenomyces interdigitalis*. Dans nos expériences, toutes le 3 sortes de kératine employées (cheveux, laine, corne de boeuf) se sont comportées de la même façon.

Pour apprécier l'influence des variations de la température et du pH sur la digestion de la kératine par les dermatophytes, nous nous sommes limités, dans nos expériences, au *Microsporum gypseum*.

L'influence de la température, sur la digestion de la kératine, a été mise en évidence de la manière suivante: On a préparé 18 tubes, contenant la suspension 0,02 % de laine pulvérisée et étagée comme cidessus. Les tubes ont été répartis en 3 groupes égaux. Les 5 premiers tubes de chacun de ces groupes ont été ensemencés, le 6-ème devant servir comme témoin. Un premier groupe a été maintenu à 20° C, un autre à 28° C, et le dernier à 37° C. Chose curieuse, les tubes maintenus à 37° n'ont présenté aucune modification visible macroscopiquement, ni même après 3 semaines. Par contre, une partie des tubes maintenus à 20° et à 28° ont montré une disparition complète des particules en suspension à l'étage inférieur, non invadé par la culture. Plus précisément, 2 tubes maintenus à 20° C ont montré cette disparition après 15 jours, tandis que dans 3 tubes maintenus à 28° C, ce phénomène s'est produit dès le 10-ème jour. La température optime paraît être donc proche de 28°.

---

[1]) Les modèles expérimentaux, ci-dessus décrits, pourraient prêter à l' erreur à cause de la chûteau fond des tubes, des particules qui forment la suspension. Par la suite, la suspension devient limpide d'elle-même. Afin d'éviter cette cause d'erreur, il faut, avant de procéder à la lecture des résultats, rouler les tubes entre les mains, obligeant ainsi les particules non digérées, à monter dans les tubes et à devenir ainsi visibles.

Dans le but d'apprécier l'influence du pH, sur la digestion de la kératine, on a préparé 18 tubes à suspension de laine pulvérisée. Ces tubes ont été répartis en 6 groupes à 3 tubes chacun. A l'aide de systhèmes tampon à phosphates, le pH a été fixé, pour chacun des 6 groupes, aux valeurs suivantes: 4,9; 5,9; 6,4; 7,1; 7,6; 8,1. La lyse de la kératine s'est produite, le plus rapidement, dans les tubes à pH = 7,6 et à pH = 8,1.

OBSERVATIONS PRÉLIMINAIRES SUR LE MODE D'ACTION DE L'ENZYME KÉRATINOLYTIQUE, PRODUITE PAR LES DERMATOPHYTES.

Bien qu'il fût admis, en général, que la désintégration de la kératine commence par la disjonction des ponts du type cystine, qui unissent les chaînes polypeptidiques, nous n'avons pas réussi, en utilisant l'épreuve au nitro-prussiate de soude, de déceler la présence des groupements — SH, qui en auraient résulté. Nous avons essayé l'épreuve au nitro-prussiate sur des cheveux et sur des squames parasités par des dermatophytes divers et aussi sur des cheveux ou de la laine invadés in vitro par ces champignons. Ce contrôle a été fait à des stades divers de l'invasion. Les résultats ont été constamment négatifs. Les mêmes résultats négatifs nous les avons obtenu avec les suspensions de kératine lysées indirectement dans le modèle expérimental décrit plus haut.

Comme témoin, nous nous sommes servis de la kératine (cheveux et squames) soumise à l'action de la potasse caustique. Celle-ci, étant une base énergique, réduit les ponts à forme — S — S — (qui unissent les chaînes polypeptidiques) élibérant ainsi les groupements — S H (9). Pour la kératine ainsi traitée, l'épreuve au nitroprussiate a été fortement positive.

Si la présence du groupement — S H, dans les lysats de la kératine, n'a pu être point décelée, en revanche, par la méthode classique, à l'aide du réactif d'ABDERHALDEN (1) (solution alcoolique 5 % d'acide picrique et solution aqueuse 10 % d'hydrate de soude, parties égales) nous y avons mis en évidence la présence des noyaux cycliques dicétopipéraziniques. Cette réaction, d'abord négative, est devenue petit à petit positive, pour redevenit négative quand l'action du dermatophyte se prolongeait.

Quant à la composition des lysats de kératine, présents dans le compartiment inférieur de nos tubes étagés, les recherches préliminaires, effectuées par nous à l'aide de la méthode chromatographique, y ont mis en évidence les amino-acides suivants: cystine, lysine, acide glutamique, alanine, leucine et valine. Cela veut dire que la digestion de la kératine est allée assez loin.

DISCUSSION

La disparition des particules de kératine contenues dans le compartiment inférieur de nos tubes étagés, c'est à dire là où les éléments figurés du dermatophyte étaient absents, ne peut être expliquée que par l'action des produits solubles, élaborés par ces dermatophytes. Ces produits ont traversé aisément les bouchons d'ouate et, parmi eux, nous sommes obligés de l'admettre, il y a eu une enzyme ou un système enzymatique, à propriétés kératinolytiques. Selon nous, le bouchon d'ouate présente deux avantages sur les filtre poreux ordinaires. Premièrement, d'après ce que l'on sait, les ferments sont des substances oxydables. Le passage des liquides, à travers les filtres d'ouate, se réalise plus rapidement, qu'à travers les filtres poreux habituels et, par conséquent, les substances, qui y sont dissoutes, ont plus de chances de les traverser sans être oxidées. En second lieu, les canalicules des filtres poreux ordinaires ont une surface bien plus grande que ceux d'un simple bouchon d'ouate et, par là, les chances, pour les substances dissoutes à être adsorbées par le filtre, deviennent plus nombreuses.

Il paraît probable que la fragmentation fine de la kératine, eût facilité l'action de l'enzyme, premièrement par l'agrandissement de la surface de contact de la kératine et, secondement, parce que la résistance de cette substance s'en trouva diminuée.

Comme il a été montré plus haut, dans les lysats des suspensions soumises à l'action des dermatophytes, nous avons décelé la présence des noyaux cycliques dicétopipéraziniques et, à l'aide de la méthode chromatographique, nous y avons mis en évidence la présence de certains aminoacides, propres à la kératine. On pourrait se demander si tous ces corps chimiques ne proviendraient, tout simplement, de la désintégration du champignon même et pas de celle de la kératine. D'après nous, cette hypothèse n'est pas admissible, car les filtrats des cultures des dermatophytes sur de l'eau maltosée simple, qui ne contenait aucun composé azoté, ont présenté une réaction à la ninhydrine absolument négative. Par contre, les lysats de kératine sus-mentionnés, ont montré une réaction à la ninhydrine, fortement positive, ce qui prouve que les produits azotés cités provenaient bien de la kératine et pas du champignon en voie de désintégration.

RÉSUMÉ

Les auteurs exposent les résultats de leurs recherches concernant l'existence, chez les dermatophytes, d'une exoenzyme capable de digérer la kératine. Ces recherches ont été motivées par le fait que, malgré la kératinophilie incontestable des dermatophytes, on ne trouve pas dans la littérature de spécialité, que peu de travaux concernant ce sujet, excepté peut-être le travail de STAHL et collab. sur les ferments du *Microsporum gypseum.* En imaginant un dispositif expérimental très simple, les auteurs ont obtenu la lyse complète de la kératine, par les dermatophytes, non seulement d'une façon directe, mais aussi indirecte, ce qui prouve l'existence, chez ces champignons, d'une exoenzyme capable de digérer la kératine. D'après les constatations des auteurs, le ferment kératinolytique des dermatophytes ne provoque pas la disjonction des ponts, de forme — S — S —, qui unissent les chaînes polypeptidiques de la kératine. Son action se traduit par l'apparition et puis par la disparition, dans les lysats de la kératine, des noyaux cycliques dicétopipéraziniques et elle se poursuit jusqu'à ce que la kératine se décompose en amino-acides. Ces aminoacides ont pu être mis en évidence au moyen de la méthode chromatographique.

SUMMARY

The authors report on the results of their research concerning the existence of an exoenzym in dermatophytes able to digest keratin. These investigations were promoted by the fact that, inspite of their keratinophily, dermatophytes are almost unknown in the literature, as source of keratinolitic enzyms. Except for the work of STAHL and coll., concerning the keratinolitic enzym of *Microsporum gypseum,* this problem was little studied up to now. The authors of the present article, suggesting a very simple experimental model, demonstrated the complete digestion of keratin, by the dermatophytes, not only directly, but indirectly too, thus proving the existence, in this fungus, of an exoenzym able to decompose keratin. The authors were not able to demonstrate the disjunction of molecular bridges — S — S —, between polypeptidic chains, as caused by the ferment. Its activity manifests itself in the appearance and later disappearance of diketopiperazin rings in the products of keratin digestion. At last, keratin is decomposed into aminoacids, which could be evidenced by chromatographic method.

Bibliographie

1. ABDERHALDEN, E. Physiologisch-chemisches Praktikum. 8. Auflage 1948—1949. Frankfurt am Main. Dietrich Steinkopf. Teil 1, 17—21.

2. Balls, A. K. & Lineweawer 1939. Isolation and properties of cristaline papain. J. biol. Chem. **130:** 669.
3. Jensen, H. L. 1930. Decomposition of keratin by soil microorganisms. J. of agric. Sci. **20:** 390—398. Cité par St. Rothman (Physiology and Biochemistry of the Skin. Chicago 1954 p. 359.)
4. Kapica, L. & Blank, F. 1957. Growth of Candida albicans on keratine as sole source of nitrogen. Dermatologica (Basel) **115:** 2, 81—105.
5. Mallinckrodt-Haupt, A. 1928. Die Protease der pathogenen Hautpilze. Arch. Dermatol. Syph. **154:** 3, 493.
6. Roberts. 1889. Proteolytische Fermente. Brit. Med. J. 7 I. Cité par Bruhns & Alexander (Handbuch f. Haut u. Geschlechtskrankheiten Bd.XI, 294 Berlin 1928).
7. Rothman, St. Physiology and Biochemistry of the Skin. Chicago 1954. p. 359.
8. Stahl, W. H., Mc Que, Mandels, G. T. & Siu, G. H. Studies of the microbiological degradation of Wool. Digestion of normal and modified fibrillary proteins. Textile Res. J. **XX,** 8. 570—579.
9. Tchernov, N. V., Aronina, I. N., Gaidarov, L. P., Goevteeva, A. A., Lesitsky, J. M., Mihailov, N. A., Stratov, I. P., Sestakova, I. S. Tehnologia Koji. Moscou Guizlerprom. 1952.
10. Truffi, M., Fermentbildung bei den Dermatophyten nebst Pigmentbildung Boll. chem. farmaceut. Sept. 1901 et Soc. med. chirurg. di Pavia 31. 3. 1905. Cité par Bruhns & Alexander (Handbuch f. Haut u. Geschlechtskrankheiten Bd.XI, 297, Berlin 1928).
11. Vanbreuseghem, R. 1953. Les champignons kératinophyles et kératinolytiques. Atti VI Congr. Int. Microbiol. Vol. V, 236, Roma 6—12 sett. 1953. Sezione XIV Miceti.

DE L'ORIGINE SAPROPHYTIQUE, TELLURIQUE DES DERMATOPHYTES; UNE ESPÈCE DU GENRE ALEURISMA, SAPROPHYTE DU SOL ET AGENT D'UNE DERMATOMYCOSE EXPÉRIMENTALE

par

R. Evolceanu & I. Alteras

(*Section Dermatologique de l'Institut de Thérapeutique de Bucarest*
Chef: *Prof.* St. G. Nicolau, *membre de l'Académie Roumaine*)

(30.VIII.1959)

D'après ce que l'on sait, les dermatomycoses superficielles ne sont pas toujours provoquées par les dermatophytes appartenant au genre *Trichophyton* et aux genres voisins. Outre ces champignons et outre les agents des candidoses superficielles et ceux du pytiriasis versicolor, de l'erytrasma etc., on trouve, parmi les agents des mycoses superficielles, exceptionellement il est vrai, des champignons de la famille des Aleurismées. Ces derniers sont des Hyphomycètes très répandues dans la nature. Les rares dermatomycoses à Aleurismées revêtent plutôt l'aspect d'une affection erythémato-squameuse, capable parfois d'imiter à s'y méprendre, les dermatomycoses communes. Le tableau clinique est alors celui d'un herpès circiné typique, à plaques érythémato-squameuses ou érythémato-vésiculeuses, circulaires, dont le pourtour est plus actif et nettement délimité, tandis que la partie centrale a une tendance à la guérison. Si ces plaques occupent les régions velues, elles peuvent provoquer la chute des cheveux ou des poils et, de cette façon, imitent, mieux encore, les dermatomycoses communes. Ainsi, à l'Académie de Médecine de Paris, en 1949, A. et R. Sartory, Meyer & Altweg (6) ont présenté un petit malade, âgé de 5½ ans, dont le torse et le cou étaient couverts de vastes plaques d'herpès circiné, tandis qu'au cuir chevelu, on voyait des plaques squameuses étendues, où les cheveux étaient, en partie, tombés. Les squames contenaient des nombreux filaments mycéliens et, après leur ensemencement, on obtint constamment une espèce d'*Aleurisma*.

Une réelle similitude entre les caractères pathogéniques des dermatophytes communs et ceux de certaines Aléurismées ne peut donc être contestée. Mais ces 2 groupes de champignons ont encore un autre caractère commun, à savoir la présence des aleuries. C'est grâce à ces spores que les cultures des dermatophytes, ainsi que celles des Aleurismées, ont souvent un aspect poudreux caractéristique.

C'est la ressemblance mentionnée, des caractères pathogéniques et botaniques, entre les dermatophytes et les Aleurismées d'une part, et, d'autre part, la présence ubiquitaire, dans les couches superficielles du sol, de certains dermatophytes (notamment le *Microsporum gypseum* et le *Keratinomyces ajelloi*) (1), qui ont inspiré l'objet de notre article.

Afin d'isoler les Aleurismées des couches superficielles du sol, nous avons eu recours au procédé, presque devenu classique, de VANBREUSEGHEM (9). En mettant des cheveux en contact avec des diverses souches de terre, récoltées tant à Bucarest qu' en province, nous avons réussi à isoler un champignon qui, d'après ses caractères botaniques, doit être rattaché au genre *Aleurisma* — LINK.

ÉTUDE MYCOLOGIQUE DU CHAMPIGNON ISOLÉ PAR NOUS. CULTURES.

Sur le milieu d'épreuve de Sabouraud, notre champignon se développe assez bien. Des le 4-ème jour, on y observe un petit duvet blanc qui, petit à petit, les jours suivants, s'agrandit (fig. 1) et, en même temps, devient poudreux. Les cultures imitent alors celles du *Ctenomyces mentagrophytes*, mais la ,,poudre'' en est un peu plus blanche et le développement un peu plus lent. Les cultures vieilles de plus d'un mois, présentent fréquemment une couleur jaunâtre assez caractéristique.

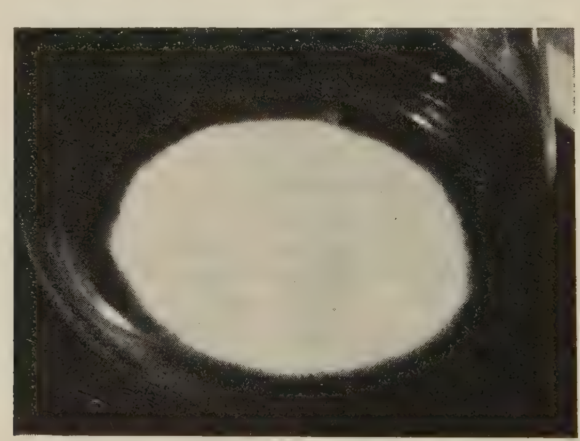


Fig. 1. *Aleurisma* - culture de 5 jours sur millieu d'épreuve de Sabouraud, en matras d'Erlenmeyer.

Sur le milieu à pommes de terre et carottes (pommes de terre, carottes et gélose à 2 %), l'aspect des cultures est le même, mais leur développement y est plus lent.

Cultures en goutte pendante.

Les filaments mycéliens paraissent assez minces (2—3 μ), ramifiés à angle droit, d'abord non cloisonnés, puis divisés par des septums en compartiments dont la longueur dépasse de 8—10 fois leur largeur. Les articles, qui résultent du cloisonnement, finissent par se séparer, en même temps que leur poutour s'arrondit. Par endroits, les rameaux mycéliens sont très allongés et à bout effilé.

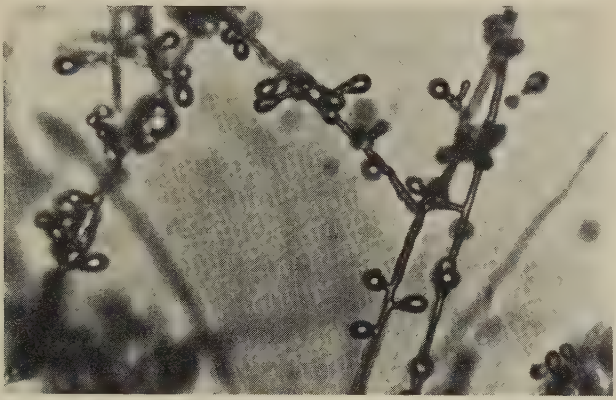


Fig. 2. Aleuries à parois épaisses (microculture de 2 semaines)

Les aleuries ont 5—6 μ de diamètre. Elles sont nettement piriformes, à parois assez épaisses (fig. 2) et s'attachent aux rameaux mycéliens, soit directement, soit à l'aide d'un court stype. On en voit aussi aux extrémités des certaines branches mycéliennes, dont la longueur est extrêmement variable. Par places, les aleuries sont très nombreuses mais leur distribution sur les rameaux mycéliens est inégale et on n'y trouve pas des branches qu'on pourrait considérer comme des sporophores vrais.

Sur le milieu à pommes de terre et carottes, les filaments mycéliens sont plus minces et moins cloisonnés et leurs articles ne se séparent pas.

Les aleuries sont moins nombreuses mais, en revanche, on y trouve des chlamydospores terminales, à parois épaisses (des chlamydospores vraies, (fig. 3), pas de simples cellules dilatées par l'epictese, comme on en observe quand les conditions deviennent peu favorables au développement des cultures). On y observe aussi des spirales molles.

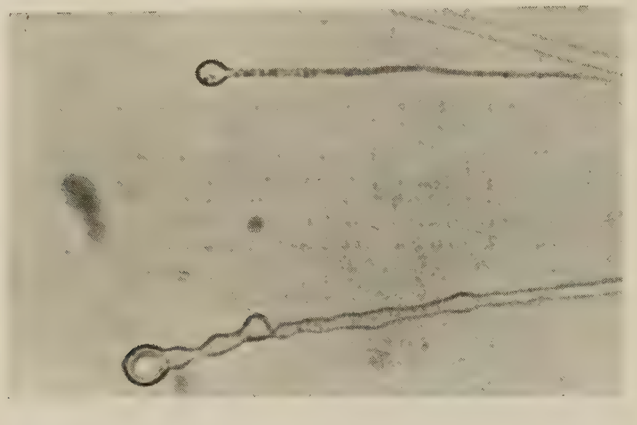


Fig. 3. Chlamydospores terminales (microculture)

Sur gélatine 2 %, après 10 jours, colonies circulaires, en cocarde à bandes concentriques, alternantes, de couleur rose et blanche. L'image microscopique rappelle celle des cultures sur milieu de Sabouraud.

Fermentations

Sous l'action de notre *Aleurisma*, la liquéfaction de la gélatine devient visible dès le 3-ème jour et la coagulation du lait dès le second. Ensemencée sur des cheveux isolés en chambre humide, l'*Aleurisma* ne se développe pas et ne provoque pas leur digestion, contrairement aux dermatophytes vivaces (*Ctenomyces mentagrophytes*, *Microsporum lanosum*, *Microsporum gypseum* etc.). L'invasion des cheveux, in vitro, ne se produit que si ceux-ci sont mis en contact avec de la terre.

Essais d'identification

D'après ses caractères botaniques, notre champignon doit être placé dans la sous-famille des Aleurismées — VUILLEMIN (10) — à aleuries monocellulaires —. C'est juste, car les spores de ce champignon restent sur place jusqu'à la disparition du mycélium et, par conséquent, doivent être considérées comme aleuries, car elles diffèrent des autres types de conidies qui se détachent dès qu'ils se sont formés.

Les caractères macro- et microscopiques des cultures appartiennent au genre *Aleurisma* — LINK 1809 (2; 4), VUILLEMIN 1910 emend. (mycélium cloisonné, très ramifié, sans conidiophores. Rameaux terminés par des aleuries ovales, à parois épaisses, non caduques, mises en liberté par la destruction du mycélium. Aspect

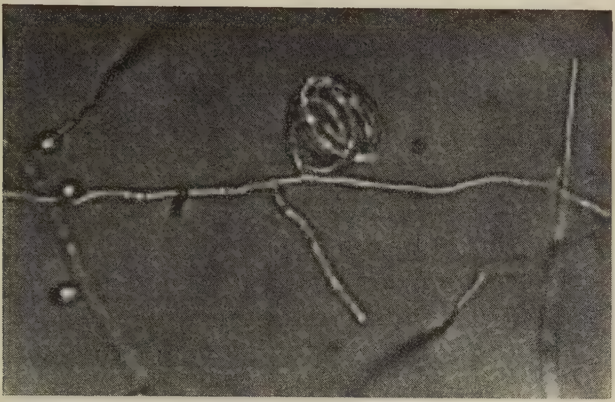


Fig. 4. Spirales molles (microculture de 35 jours)

farineux des cultures). Morphologiquement il est prôche à l'*Aleurisma lugdunense* — Vuillemin 1924 (3; 5) (aleuries sessiles ou pédiculées, latérales ou terminales, chlamydospores présentes). Il en diffère cependant par le manque du pigment dans les cultures.

Inocculations expérimentales

Des inoculations expérimentales à l'homme ont réussi, bien que difficilement. Sur huit personnes, âgées de 15 à 40 ans, hommes ou femmes, qui ont été inoculées à l'avant-bras, une seule s'est montrée réceptive envers notre champignon. C'était une femme âgée de 36 ans. Six jours environ après l'inoculation, il s'est développée, au point inoculé, une tache rouge, nettement ovalaire, bien délimitée (fig. 5), dont le grand diamètre mesurait approximativement 2 cm et dont le pourtour était marqué par une couronne de petites vésicules. La lésion a persisté, comme telle, 5 jours, après quoi elle a commencé à guérir. Pendant tout ce temps, on a trouvé de filaments mycéliens dans les vésicules et squames qui, ensemencées, ont reproduit la culture inoculée.

Les inoculations à l'animal (8 souris blanches et 4 cobayes) sont restées sans effet.

Propriétés immuno-biologiques

Avec les cultures de l'*Aleurisma* isolée par nous, nous avons préparé un extrait spécifique (analogue à la trichophytine), d'après une méthode ¹) utilisée en bactériologie pour obtenir les toxines

---

¹) La masse mycélienne de cultures vieilles de 5 à 6 semaines, est lavée, puis deshydratée par le maintien en acétone 24 heures, séchée et finement triturée. La poudre qui en résulte est délayée dans de l'eau distilée, en proportion de 1 à 9, maintenue à 80° C pendant une heure et centrifugée. Le surnatant est soumis à la dialyse à l' eau courante, pendant 2 jours, puis à l'eau distilée et, enfin, séché. Dans notre service on emploie souvent ce procédé pour préparer la trichophytine.

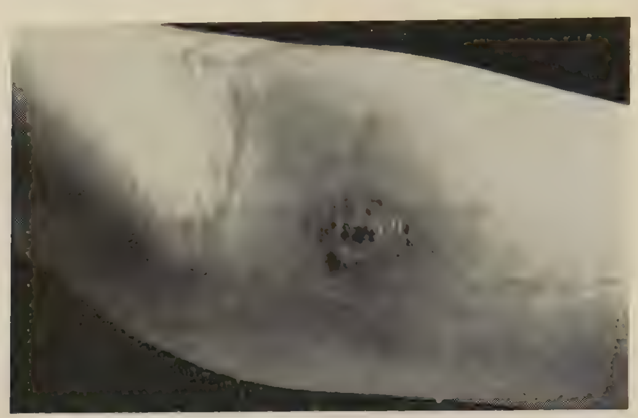


Fig. 5. Mycose expérimentale humaine à Aleurisma (Face antérieure de l'avant-bras).

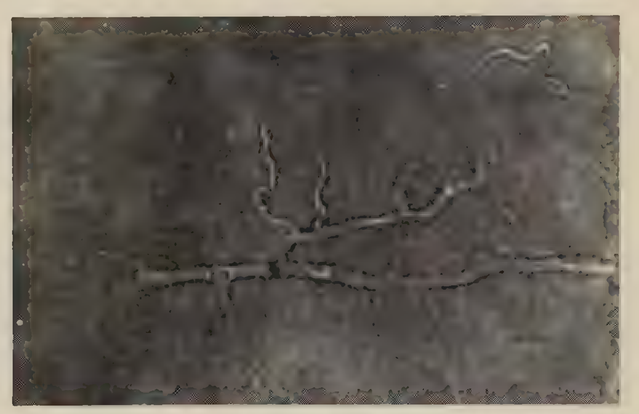


Fig. 6. Filaments mycéliens ramifiés, dans les squames de la lésion expérimentale.

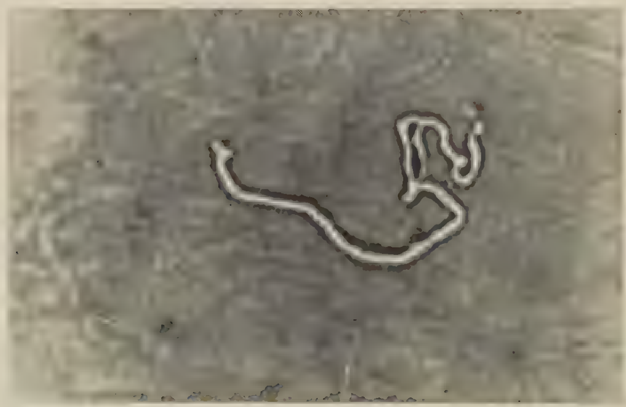


Fig. 7. Détail de la figure précédente.

microbiennes. Douze malades, atteints de diverses mycoses inflammatoires, provoquées par le *Ctenomyces mentagrophytes* ou par le *Ctenomyces interdigitalis*, ont été inoculés dans le derme, avec cet extrait. Parallèlement, on leur a inoculé, toujours dans le derme, de la trichophytine. Voici les résultats de ces intradermo-réactions:

Diagnostique	*Réactions à la trichophytine*	*Nombre des malades*	*Réactions à l'aleurismine*	*Nombre des malades*
1. Kérion de Celse (cuir chevelu ou barbe)	+ + +	3	+	3
	+ +	4	+	1
			—	3
	+	2	—	2
2. Epidermophytie inflammatoire	+ +	2	+	1
			—	1
	+	1	+	1

Le tableau ci-dessus nous montre que l'intradermo-réaction à l'aleurismine a été, chez tous les malades, beaucoup moins forte que l'intradermo-réaction à la trichophytine, pratiquée en même temps. Les réactions faiblement positives à l'aleurismine ont coincidé généralement avec des réactions fortement positives à la trichophytine et les réactions négatives à l'aleurismine ont coincidé avec des réactions moyennement ou faiblement positives à la trichophytine. Un seul cas a fait exception à cette règle, à savoir un malade atteint d'épidermophytie inflammatoire, chez lequel l'intradermo-réaction à l'aleurismine, ainsi que celle à la trichophytine, ont été toutes les deux, faiblement positives. Quoique le nombre des sujets, sur lesquels nous avons pratiqué les intradermo-réactions, fût modeste, il n'en résulte pas moins qu' entre l'aleurismine et la trichophytine il existe un certain parallélisme immuno-biologique.

DISCUSSION

Notre champignon, nous l'avons isolé du sol, à l'aide du procédé de VANBREUSEGHEM, procédé qui permet d'isoler les champignons kératinophyles. C'est vrai que la plupart des cheveux mis en contact avec de la terre, ont été envani par d'autres champignons (surtout par le *Microsporum gypseum* et la *Keratinomyces ajelloi*) mais nous avons aussi trouvé quelques cheveux attaqués et détruits par l'*Aleurisma*. Cependant, ce champignon, ensemencé sur des cheveux, isolés et suspendus en chambre humide, ne s'est pas développé et ne les a pas attaqué, tandis que, les conditions étant les mêmes, des dermatophytes vivaces comme le *Microsporum gypseum* etc., envahissent rapidement les cheveux. Cela veut dire que notre *Aleurisma* est moins kératinophyle que les sus-dits dermatophytes. On peut se demander si, mise en contact avec la terre, la kératine des cheveux n'a pas été sujette à des modifications et qu'elle n'en soit pas devenue moins résistante. Le procédé de VANBREUSEGHEM nous permet d'isoler du sol, en premier lieu, les champignons les plus kératinophyles, pas les plus répandus et, si l'*Aleurisma* est moins kératinophyle que le *Microsporum gypseum*, par exemple, nous aurons moins de chances à l'isoler, les cheveux étant immédiatement envahis par les champignons les plus kératinophyles, les autres ne pouvant plus s'y développer. Si, dans notre pays nous avons trouvé le *Microsporum gypseum*, dans le sol, absolument partout où nous l'avons cherché, tandis que l'*Aleurisma* seulement 4 fois, cela ne veut pas dire que ce dernier champignon soit moins répandu dans le sol. Tout simplement, il faut se donner plus de peine à l'isoler. Il paraît donc bien probable que l'espèce d'*Aleurisma*, isolée par nous, ne soit pas moins répandue dans le sol que le *Microsporum gypseum*.

Parmi les moisissures kératinophyles qui habitent les couches superficielles du sol, les unes comme le *Microsporum gypseum*, pénétrant accidentellement dans l'épiderme corné des animaux terrestres, se sont petit à petit adaptés à la vie parasitaire. On peut les considérer tout aussi bien des parasites que des saprophytes. Par contre, d'autres moisissures comme, par exemple, l'*Aleurisma*, moins kératinophyles, ne deviennent qu'exceptionellement des parasites de l'épiderme corné.

En ce qui concerne les espèces du genre *Aleurisma*, leur capacité d'envahir l'épiderme s'explique aussi par une certaine parenté qui existe entre ces espèces et les dermatophytes communs. Outre les affinités botaniques, dont il a été question plus haut, et outre les affinités pathogéniques et antigéniques, il y a entre ces champignons une réelle ressemblance, concernant leurs propriétés fermentatives. La capacité de liquéfier la gélatine est propre tant aux dermatophytes qu'aux espèces du genre *Aleurisma*. Elle prouve que les dermatophytes, tout aussi bien que l'*Aleurisma* (l'espèce isolée par nous et celle isolée par SARTORY et coll. (7)) possèdent un ferment analogue à la trypsine.

Bien que les espèces appartenant au genre *Aleurisma* ne comptent pas parmi les agents habituels des mycoses superficielles de la peau, il ne faut jamais les perdre complètement de vue. Outre les cas déjà publiés par des divers auteurs, nous avons eu, personnellement, l'occasion d'observer, dernièrement, une jeune femme, âgée de 20 ans, chez laquelle nous avons trouvé cette même espèce d'*Aleurisma*. Il s'agissait d'une plaque erythémato-squameuse, circulaire, ayant 2 cm de diamètre, très superficielle, localisée à la face antérieure de l'avant-bras. Les squames contenaient des filaments mycéliens et, par leur ensemencement, on obtint constamment des cultures d'*Aleurisma*. Malheureusement, cette malade n'est plus revenue et nous étions incapables de poursuivre l'évolution de la dermatose. Bien sûr qu'autrefois, en pareilles occasions, nous aurions pris ce champignon pour un contaminant banal et ne lui aurions accordé la moindre attention.

En fin de compte, l'existence, outre les dermatophytes communs, d'autres saprophytes du sol, capables de parasiter l'épiderme corné et, bien plus, la parenté qui existe entre ceux-ci et les premiers, viennent à l'appui de la thèse, d'après laquelle, les dermatophytes proviennent des champignons saprophytes du sol.

RÉSUMÉ

Les auteurs ont isolé, par le procédé de VANBREUSEGHEM, des couches superficielles du sol, un champignon appartenant au genre *Aleurisma* LINK. Ils ont réussi, bien que difficilement, de l'inoculer à l'homme, chez lequel ce champignon a provoqué un herpès circiné typique. Le rôle pathogène de l'*Aleurisma* a été confirmé par la rétroculture des vésicules de la lésion expérimentale, rétroculture qui a reproduit exactement le champignon inoculé. Outre les affinités botaniques (présence des aleuries) et pathogéniques (capacité de provoquer des dermatomycoses superficielles), les auteurs ont découvert une certaine parenté antigénique entre les dermatophytes communs et le champignon isolé par eux. Bien plus, leurs propriétés fermentatives étaient aussi semblables. La présence dans le sol de champignons autres que les dermatophytes communs, mais assez proches d'eux, tant du point de vue botanique et fermentatif que du point de vue pathogénique et antigénique, vient, selon les auteurs, à l'appui de la thése, d'après laquelle, les dermatophytes proviennent des champignons saprophytes du sol.

SUMMARY

The authors isolated, from the superficial layers of the soil, a fungus belonging to the genus *Aleurisma* — LINK. Although with difficulty, they were able to inoculate this fungus in man. The lesion it provoked, was of herpes circinatus type. The cultures obtained from the vesicles of the experimental lesion reproduced the strain first inoculated. Besides the botanical (presence of aleuries) and pathogenic affinities (capacity of causing superficial mycosis) the authors were able to demonstrate a certain antigenic resemblance between this fungus and the common ringworm agents. Their fermentative properties were also similar.

The presence in the soil of fungi related to dermatophytes, their botanical, fermentative, pathogenic and antigenic affinities are considered by the authors as an argument in behalf of the thesis, according which, dermatophytes take their origin from saprophytic fungi of the soil.

P.S. Au moment où ce travail était déjà sous presse, nous avons reçu, de la part de Mr. L. AJELLO, des extraits de quelques uns de ses travaux, parmi lesquels, un article signé par L. GEORG,

W. Kaplan, L. Ajello, W. Williamson & Ev. Tilden (J. Inv. Dermat. Vol. 32 No. 4, 1959) où ces auteurs démontrent la présence du *Keratinomyces ajelloi* comme agent d'une mycose spontannée de l'écureuil et les résultats positifs de son inoculation au cobaye. Nous sommes reconnaissants à M. Ajello et le prions de nous excuser de n'avoir pas connu cet article.

Bibliographie

1. Ajello, L. 1956. Soil as natural Reservoir for Human Pathogenic Fungi. Science **123,** 3203, 876—879.
2. Brumpt E. Précis de Parasitologie Paris, 1936 p. 1799.
3. Id., p. 1801.
4. Coudert, J. Guide pratique de Mycologie Médicale. Paris 1955 p. 277.
5. Id. p. 279.
6. Sartory, A. & R., Meyer, J. & Altweg, J. 1949. Dermatomycose trichophytiforme due à une espèce nouvelle du genre Aleurisma. Bull. Acad. Nat. Méd. **133,** 519—523.
7. Sartory, R. Meyer, J. & Malgrai, J. 1951. Bull. Acad. Nat. de Méd. **135,** 304—305.
8. Stepanistcheva Z. G. Otcherki po gribkovim zabolevaniam koji. Medguiz Moscou 1955 pp. 98—103.
9. Vanbreuseghem R. Les champignons kératinophyles et kératinolytiques. Atti VI Congr. Int. di Microbiol. Vol. V, p. 236. Roma 6—12 sett. 1953. Sezione XIV Miceti.
10. Vuillemin P. Les champignons parasites et les mycoses de l'homme. Paris 1931 p. 138.

COREMIELLA CUBOIDEA (SACC. ET ELL.) CIF. ET CAR. N. COMB. ISOLATA DA FECI UMANE

G. Caretta

Centro di Micologia Medica presso l'Istituto Botanico dell' Università di Pavia (Direttore: Prof. R. Ciferri)

Istituto di Clinica Medica Generale e di Terapia Clinica dell' Università di Pavia (Direttore: Prof. P. Introzzi)

(27.VII.1959)

In una precedente contribuzione allo studio morfologico di specie del genere *Geotrichum* (Caretta, 1959) anticipammo la notizia che, accanto a ceppi riferibili con sicurezza al *G. candidum* Link, erano stati isolati anche forme che avevano affinità con *Coremiella* Bubák et Krieger (in Bubák, 1912).

In questa contribuzione diamo conto dei risultati dello studio di questi funghi affini e mal differenziati da *Geotrichum* quando invece, secondo la nostra opinione, meritano di essere distinti anche come genere.

Origine dei ceppi

I ceppi di cui alla presente contribuzione, in numero di quattro e segnalati col numero romano dal I al IV, sono stati isolati dalle feci di 130 ammalati ricoverati in Clinica Medica affetti da affezioni diverse, nel corso di ricerche svolte ad altri fini.

Premettiamo subito che mai ci è dato osservare un qualsiasi rapporto tra le affezioni degli ammalati e la presenza di questi ceppi. Gli isolamenti culturali venivano praticati simultaneamente per ogni ammalato dall'espettorato e dalle feci. Contrariamente a quanto si è riscontrato fra i lieviti, nessuno dei ceppi in istudio è stato isolato, oltrechè nelle feci, pure dall'espettorato dello stesso ammalato. Solo in un caso è stato isolato simultaneamente nelle feci un lievito (e precisamente una *Candida albicans*) e uno dei ceppi qui studiati.

Metodi di studio

I metodi di studio sono quelli stessi impiegati dai ricercatori per lo studio dei lieviti e in ciò ci siamo attenuti alle tecniche di Car-

MICHAEL (1957), CIFERRI (1958), LODDER & KREGER VAN RIJ (1952), ecc. Sebbene la cultura sia agevole in tutti gli usuali mezzi di laboratorio, ci è parso particolarmente utile, come per lo studio di *Geotrichum*, l'impiego di un mezzo relativamente povero di sostanze nutritive, quale è l'agar all'infuso di farina di mais, lo stesso impiegato per provocare la formazione di clamidospore in *C. albicans*. I vantaggi di questo terreno su quelli più ricchi in metaboliti sta nello schematizzare, per così dire, la morfologia microscopica del fungo, provocando la genesi degli elementi morfologici compatibili con l'attività vitale del fungo e utile al suo studio. Agli stessi fini risponde il terreno all'acetato sodico agarizzato impiegato per provocare la sporificazione dei lieviti sporigeni. Lo studio morfologico e culturale è pure stato integrato dall'esame comparativo su terreni ricchi quali l'agar di Sabouraud glucosato. Per i terreni liquidi ci siamo limitati allo studio della crescita del fungo indagandone le caratteristiche culturali e morfologiche del velo in acqua pepto-glucosata.

Per quanto concerne le indagini sull'attività biochimica sono state eseguite le seguenti ricerche: 1) prove dell'attività fermentativa 2) prove di assimilazione dei glucidi e dei nitrati.

Per maggiori dettagli si veda il nostro studio su *Geotrichum*.

MORFOLOGIA DEI CEPPI STUDIATI SU TERRENI SINTETICI

Caratteri culturali.

Terreni solidi (agar Sabouraud ordinario). Colonia gigante dopo 15 giorni a temp. ambiente (20—22° C) rotonda, delle dimensioni di mm 10—15 circa di diam., densa, di color crema; la parte centrale è sopraelevata, finemente verrucoide e vermicolata. La colonia degrada lentamente alla periferia irregolarmente striata nel senso del raggio sino ai margini sottili e frangiati, dove sono maggiormente visibili zonature concentriche.

Dei quattro ceppi isolati il I, II, III hanno le caratteristiche suindicate. Il IV se ne differenzia poichè, a pari durata di cultura, la colonia è più sviluppata (diam. 25—30 mm, contro 10—15 mm). La forma cerebroide vermiculata è limitata soltanto alla parte centrale e le ondulazioni sono più confuse e irregolari. Il restante della colonia è pianeggiante, quasi liscio, con un margine irregolarmente festonato e plumosetto. Vi si accennano a sopracrescite in due ben delimitate parti della colonia che hanno l'aspetto di variazioni settoriali o di modificazioni della fase R o della fase S. Terreno all'infuso di mais agarizzato; colonia piana sottile d'aspetto quasi pelliculoso, incolore, estesa (diam. 40 mm) con margine indistinto: è visibile la crescita raggiata del micelio.

Poichè per quanto riguarda la morfologia microscopica non si hanno differenze tra i ceppi corrispondenti all'uno e all'altro tipo di colonia, non teniamo in conto queste modificazioni di forma delle colonie, probabilmente dovute ad un diverso sviluppo delle ife approfondentesi nel terreno di crescita, nel senso che nelle colonie più piccole e più tormentate si è avuto uno sviluppo in profondità maggiore di quanto non si sia avuto nella colonia relativamente pianeggiante, a sviluppo del micelio prevalentemente in superficie.

CARATTERI MORFOLOGICI

Avendo i quattro ceppi, come si è detto in precedenza, un'identica morfologia, la descriviamo unitariamente. L'osservazione microscopica viene effettuata dopo 1, 2, 5 giorni di crescita. Le culture sono montate in lattofenolo o lattofucsina o trypan-blu lattico.

Terreno Sabouraud peptoglucosato.

Coloniette (osservate nelle culture secondo il metodo di RIVALIER-SEYDEL) a sviluppo raggiato con ife irradiantesi dal centro, lunghe sino a 350—450μ.

Le ife miceliche originano da talli basali ovoidali del diametro massimo di 9,6μ o da talli rettangolari (12 × 6,2μ) a parete spessa e bene evidente contenente citoplasma omogeneo.

Il micelio, settato, è composto da tratti miceliari jalini (3,6—7,2 × 3,6—2,4μ) a contenuto citoplasmatico granuloso, irregolarmente distribuito talora quasi vuoti di contenuto citoplasmatico e con pareti laterali in collasso, specie nel tratto medio. Tali tratti miceliari jalini sono intercalati da cellule del micelio più corte, a contenuto citoplasmatico denso e opaco, di forma ora rotonda (diam. 4,8—3,6μ) ora ovalare, ora cuboidea (7,2 × 3,6μ) ora a barile (4,8μ di larghezza × 3,6μ di lunghezza). Queste cellule miceliari opache possono essere intercalate isolatamente a tratti miceliari jalini, o possono essere seriate in numero vario, da 3—7 a più.

La settazione dei tratti miceliari jalini e delle formazioni opache delle ife è rettilinea, e perpendicolare all'asse più lungo dell'ifa. Allorchè sono presenti i rami laterali l'ifa primaria può vuotarsi del contenuto citoplasmatico, dando origine a un tratto jalino e trasparente od alle porzioni opache dell'ifa sopradescritte, maggiormente da queste ultime. I rami sono composti ora da cellule rotondeggianti opache (4,8μ di diam.) seriate, ora da cellule cuboidee opache (3,6 × 2,4μ) in numero vario da 5 a 15. Talora i rami laterali sono formati da tratti miceliari jalini con pareti sottili (1,6μ di spessore) terminanti con cellule acrogene rotonde od ovalari, opache, del diam. di 4,8μ.

La parte terminale dell'ifa è composta frequentemente da cellule cuboidee opache o a sezione parallelepipeda, opache (4,8 × 3,6μ) a pareti spesse e bene evidenti da cui originano rami laterali brevi e tozzi, composti da 2—7 cellule opache, sì da dare all'ifa un aspetto simile a ramificazione a corna di cervo.

Terreno all'infuso di mais agarizzato

Crescita della colonia di tipo raggiato. Da un tallo basale ovoidale del diam. di 4,8μ circa, si diparte un micelio settato dello spessore di 3,6μ, jalino con ife composte da tratti miceliari jalini (48—7,2 × 4,8μ), che si ramificano in rami laterali ora pure jalini; ora composte da tratti miceliari a contenuto citoplasmatico più denso e opaco. Tali tratti miceliari a contenuto citoplasmatico denso e opaco, alla colorazione con Trypan-blu presentano una colorazione in parte brunastra e in parte bluastra. La loro conformazione è per la maggior parte rettangolare (4,8—3,6 × 2,4—1,2μ) o cuboidea, ma può essere anche rotondeggiante (diam. 4,8μ). Tali cellule sono intercalate a tratti miceliari jalini in numero vario da 2 a 10, seriate, con settazioni rettilinee e sono rivestiti da una spessa tunica bene evidente. Frequentemente sono visibili piccolissime appendici triangolari in coorispondenza degli angoli delle artrospore. Ogni cellula contiene uno o due nuclei.

I rami laterali originano sia da tali cellule miceliche a contenuto citoplasmatico che permarranno opache o che diverranno jaline e in quest'ultimo caso appena sotto la estremità distale del tratto miceliare. Questi rami possono essere costituiti o da soli tratti miceliari opachi, o da tratti miceliari jalini. Quasi sempre la parte distale di ogni ramo laterale termina con formazioni cellulari opache in numero vario da 1 a 7 e più. Frequenti sono le ramificazioni a tridente dei tratti miceliari jalini. Alla estremità libera generalmente delle ramificazioni laterali raccorciate, possono vedersi cellule rotondeggianti e raccorciate che, nei casi estremi, somigliano a clamidospore sessili non molto spessamente tunicate, ma a contenuto citoplasmatico opaco e ben colorato. Più spesso di ha una serie di cellule transizionali ed intermediarie tra quelle usuali del micelio e queste ultime, vale a dire gradualmente raccorciate e allargate, della qual catenella quella acrogena estrema tende alla forma sferica. In qualche caso tale seriazione è più irregolare o meno manifesta. Non frequentemente queste catenelle monilioidi, anzichè apicali sono intercalari. Interpretiamo questi elementi come degli abbozzi di clamidospore con caratteri di ipnospore. Nella parte distale dell'ifa micelica i tratti di micelio jalino aumentano di spessore (4,8μ) mentre gli elementi opachi seriati assumono un aspetto irregolare ora triangolare, ora trapezoide (7,2 × 4,8μ), generandosi in file adiacenti a penicillio e appiattendosi talchè i singoli elementi che costituiscono le file penicillate apicali, pur conservando ancora alcuni l'aspetto cuboideo o rettangolare, presentano la base di dimensioni superiori all'altezza. Il concrescere di queste porzioni apicali trasversalmente settate e l'accostarsi in parallelo l'una all'altra, finisce per formare degli aggregati vagamente dendroidi e brevemente, ma tozzamente, ramosi, più espansi per un maggior diametro alla sommità e più ristretti verso l'inserzione dell'ifa madre, sicchè nel loro insieme hanno un aspetto che varia dal cilindrico-ovato e irregolarmente clavato. Sono queste formazioni quelle che consideriamo di tipo coremioide reptante e che a nostro giudizio sono caratteristiche del genere *Coremiella* di cui si dirà più oltre. In vecchie culture il quadro è più confuso per il coalescere di numerosi

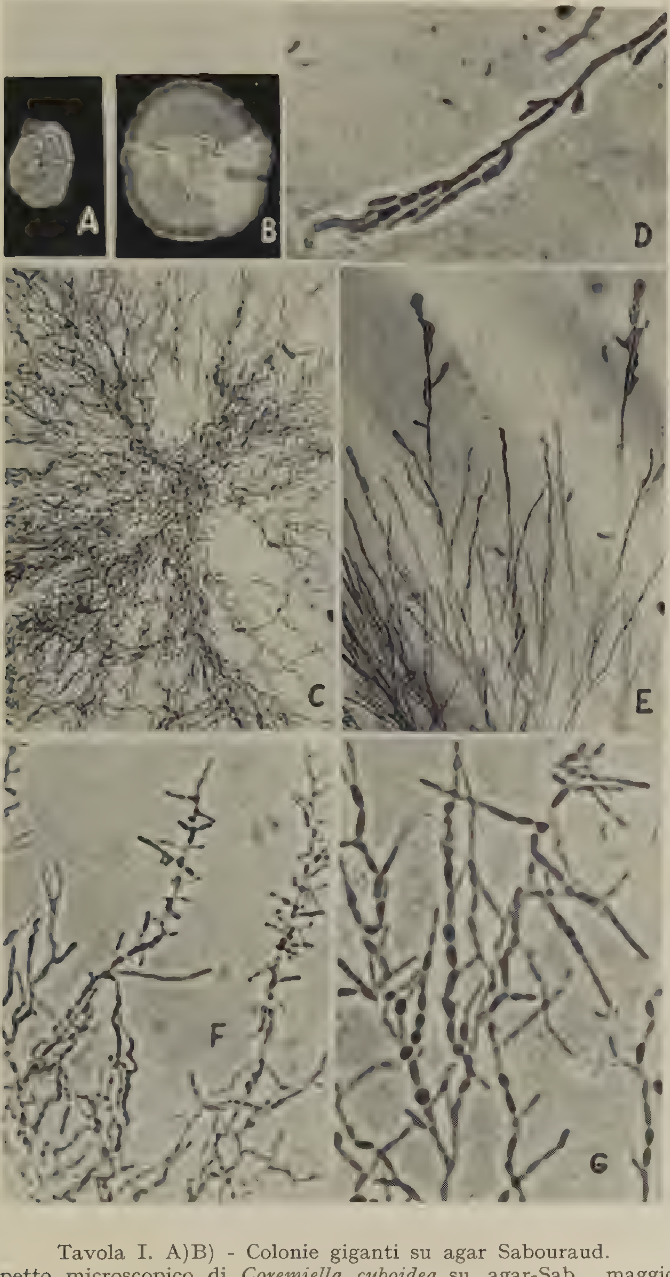


Tavola I. A)B) - Colonie giganti su agar Sabouraud.
C) - Aspetto microscopico di *Coremiella cuboidea* su agar-Sab., maggiormente evidenziato in F)G).
E) - Aspetto microscopico su agar-mais; D) -particolare di E).

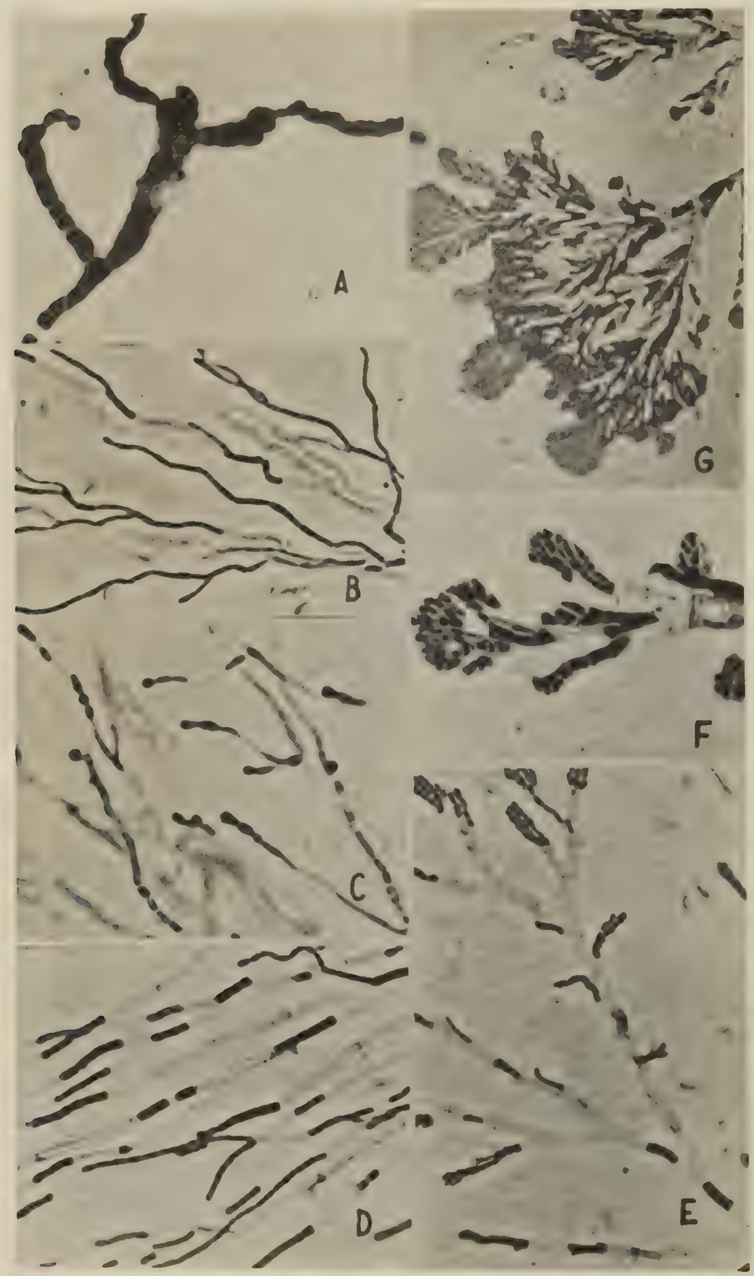


Tavola II B)D) - Aspetto microscopico di *Coremiella cuboidea* su agar-mais: sono evidenti i tratti miceliari jalini intercalati da cellule opache.

C) - *Coremiella cuboidea* su agar-mais, cellule miceliche clamidosporo - simili apicali e intercalari.

A)E)F)G) - Fasi di sviluppo delle formazioni apicali miceliche di tipo coremioide su agar-mais.

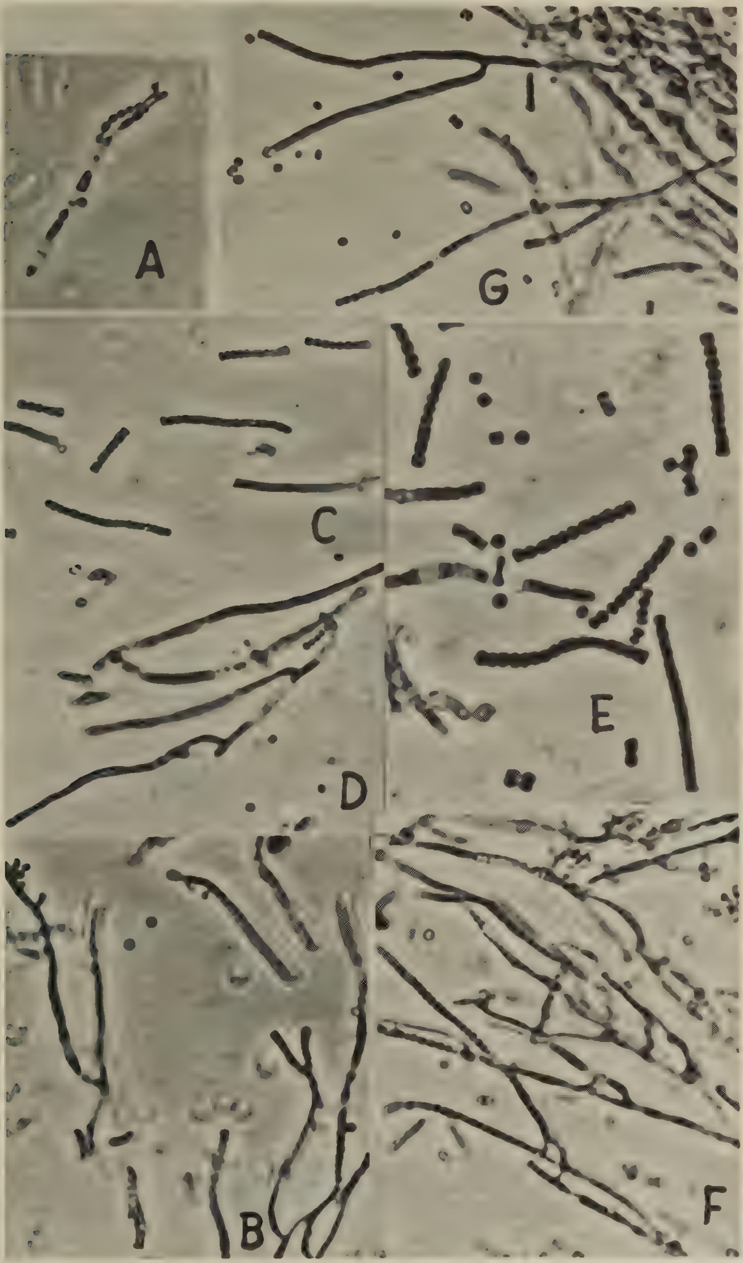


Tavola III. Aspetto microscopico dell'esemplare autentico di *Oospora cuboidea*.

rami penicillioidi, sì da formare un piano di ife confusamente trasparenti od opache con aspetto penicillioide divaricato. Si osservi che queste formazioni mancano assolutamente nelle forme del genere *Geotrichum*.

Terreno all'acetato sodico agarizzato

All'osservazione microscopica su lamina le coloniette presentano aspetto ramoso a sviluppo irregolare con ife tortuose della lunghezza di 300μ circa. Le ife miceliche originano da talli basali ovoidali di 4,8μ circa di diam., a parete bene evidente. Il micelio è formato da ife settate con segmenti miceliari di 12—7,2 × 2,4μ (lunghezza massima μ 36), a contenuto citoplasmatico granuloso più o meno abbondante e da tratti miceliari opachi seriati, in numero vario, di forma ora rettangolare di 48—7,2 × 2,4μ, ora cuboidea di 4,8 × 2,4μ, ora ovalare, ora rotondeggiante del diam. 3,6μ. I rami laterali sono brevi e nella parte terminale dell'ifa tozzi e ridotti a 1—3 elementi. Questi originano da tratti miceliari a scarso contenuto citoplasmatico e da tratti miceliari a contenuto citoplasmatico più denso. Sono formati in genere da tratti miceliari rettangolari di 4,8 × 2,4μ, talora ovalari del diam. di 3,6μ, seriati e che terminano talora con una formazione acrogena rotondeggiante del diametro di 4,8μ, simili a quelle particolari clamidospore descritte in precedenza.

Caratteri biochimici

Fermentazione: i risultati sono stati uniformi per tutti i ceppi studiati. Come in precedenza le fermentazioni sono state eseguite secondo il metodo Durham in acqua all' 1 % di peptone e con 1 % di glucidi a temperatura ambiente. Lettura sino ad un massimo di 10 giorni.

Glucide	*Gas*	*Acido*
Glucosio	—	+
Galattosio	—	+
Lattosio	—	—
Saccarosio	—	—
Raffinosio	—	—
Maltosio	—	—

Assimilazione degli idrati di carbonio e dei nitrati: le prove di assimilazione sono state giudicate dall'opacità dei mezzi liquidi di cultura in presenza di nitrato potassico come sola fonte di azoto e dei vari glucidi come sorgente di carbonio. In ciò ci siamo attenuti alle tecniche di CIFERRI (1958) e alle norme di LODDER & KREGER VAN RIJ (1952).

Glucide

Glucosio	+
Galattosio	+
Saccarosio	+
Lattosio	±
Raffinosio	+
Maltosio	+

Sostanze azotate:

Peptone	+ (testimonio)
KNO_3	±

ASPETTO MACROSCOPICO DEL FUNGO SU TERRENI NATURALI (LEGNO)

Come verrà spiegato più oltre, poichè l'*O. cuboidea* è stata descritta da SACCARDO ed ELLIS su legno di quercia, abbiamo voluto studiare le caratteristiche culturali e morfologiche dei quattro ceppi su diversi legni. Pure con riferimento a quanto si dirà più oltre a proposito della *Coremiella ulmariae* per le stesse ragioni le prove culturali sono state estese a rametti legnosi e fogliosi di *Spiraea ulmariae.* Dei rametti legnosi e fogliosi delle varie piante sono stati autoclavati in presenza di un eccesso d'acqua e quindi insemenzati con gli stipiti fungini in presenza della stessa acqua, e mantenuti a temperatura ambiente in capsule Petri per 15 giorni.

Le osservazioni sono state eseguite appunto dopo 2 settimane. Le eventuali colorazioni delle colonie si debbono a sostanze contenute nel decotto di ognuno dei legni.

Spirea. Crescita buona; il fungo si sviluppa discretamente sulla corteccia, sulle foglie e meglio sulle sezioni trasversali del legno; colonie cotonose, bianche, opache.

Salice. Crescita buona; il fungo si sviluppa discretamente sulla corteccia dove presenta un aspetto cotonoso ora bianco con ife erette di 1—2 mm ora roseo cremoso a superficie opaca o lucenti. Nelle sezioni tarsversali del legno la crescita è rigogliosa con colonie inizialmente cotonose bianche e successivamente roseo-brune cerebroidi lucenti di consistenza muco-cremosa.

Pesco. Crescita discreta sia sulla corteccia che sulle sezioni trasversali; colonie di color bianco-rosee, cotonose, ora isolate ora confluenti.

Bosso. Crescita buona sia sulla corteccia del legno che sulle sezioni trasversali; colonie di color biancastro, muco-cremose, lucenti a superficie irregolare.

Mandorlo. Crescita buona sulla corteccia ottima sulle sezioni trasversali del legno; colonie bianche o bianco-rosa, vellutate o cotonose, confluenti, opache.

Calicanto. Crescita discreta sulla corteccia, ottima sulle sezioni trasversali del legno. Colonie di colorito variabile dal verdastro al marrone chiaro, gelatinose, cerebroidi.

CARATTERI MORFOLOGICI

La crescita del fungo su salice, pesco, bosso, mandorlo, castagno non mette in evidenza dal punto di vista morfologico differenze evidenti nei riguardi del diverso substrato; in tutti è marcato notevole polimorfismo culturale riguardante principalmente gli elementi fruttiferi del fungo.

All'osservazione microscopica si rileva un micelio abbondante, settato composto da tratti ora jalini di 24 × 3,6μ, ora a citoplasma granuloso, irregolarmente distribuito. Altri tratti hanno ife settate composte da tratti miceliari jalini di 9,6 × 2,4μ e da tratti di micelio opaco di 6 × 2,4μ. Frequentemente sono osservabili tratti di micelio composti da sole artrospore opache, rettangolari, delle dimensioni di 6 × 2,4μ che successivamente assumono un aspetto cuboideo o subcuboideo. Come nelle culture su Sabouraud si hanno cellule rotondeggianti opache, seriate o isolate di un diametro variante da 4,8 a 7,2μ, sono intercalate da tratti di micelio. Tali cellule opache e rotondeggianti non sono gemmanti. In alcuni punti della porzione apicale dell'ifa sono presenti artrospore cuboidee opache e seriate di 4,8 × 3,6μ, che ricordano molto la porzione iniziale delle formazioni coremioidi descritte in agar-mais.

L'aspetto morfologico del fungo su *Spirea* è più uniforme e si avvicina molto alla morfologia di *Oospora cuboidea*. Le ife settate sono composte da tratti di micelio vacuo e trasparente, o a contenuto citoplasmatico granuloso, di 36—4,8 × 2,4μ e da artrospore opache rettangolari di 4,8 × 2,4μ o cuboidee di 2,4μ circa. I rami sono composti da artrospore seriate rettangolari alla base cuboidee nella porzione mediana e ovoidali dal diametro di 2,4μ nella porzione terminale.

Con ciò si confermano le osservazioni effettuate ex natura da McWEENEY (1895) e nello stesso tempo la similarità con *Oospora cuboidea* di cui si dirà più oltre.

SIGNIFICATO ED EVOLUZIONE DEGLI ELEMENTI MORFOLOGICI

Il processo di artrosporulazione è un processo endogeno quale è stato descritto da HUGHES (1953) e confermato da TUBAKI (1958), e avviene all'incirca con le stesse modalità iniziali osservate in *Geotrichum*. Ma la differenziazione delle endospore è quivi completa mentre ancora si trovano racchiuse in seno all'ifa materna, dalla quale si liberano per collasso, delle pareti di questa. La presenza o meno di piccolissime appendici jaline, che rappresentano il residuo dell'ifa madre, si ha soltanto in quanto le artrospore seriate non si formano in file consecutive nelle quali l'una sia strettamente adiacente all'atra. Se le artrospore sono continuativamente seriate, la traccia dell'ifa materna è quella di una sottile tunica esterna, più o meno leggermente svasata, generalmente male evidente in quanto non si colora con i coloranti usuali. Ma infuocando a vari livelli si rivela come una sezione a menisco concavo. Se invece un tratto maggiore di micelio vacuo separa due artrospore adiacenti, la tunica di una delle due può rivelarsi come una duplice sottile proiezione dovuta ad un maggior raggio di curvatura del menisco di divisione. Nei casi fortunati in cui sia rimasto sugli orli del menisco una qualche frazione di citoplasma interstiziale questo assume una colorazione che rende evidente il frammento di membrana residuale. In proiezione ottica su un piano, al microscopio, assume l'aspetto di due piccolissimi diverticoli completamente marginali, rispetto all'artrospora contenuta nel frammento di ifa.

Per queste ragioni noi non saremmo portati a dare molto valore a questa caratteristica alla quale hanno accennato od hanno insistito vari autori (ad es. GOIDÁNICH, 1935), nei riguardi della tassonomia del fungo.

Dobbiamo accennare ad un altro aspetto della morfologia che differenzia questo fungo dai *Geotrichum*. Come si è detto in precedenza alcuni tratti di artrospore seriate in agar Sabouraud glucosato tendono ad assumere una forma ovalata, ellittica e persino sferoidea: generalmente queste forme si alternano o si susseguono ad artrospore a sezione ottica quadrata o rettangolare, ma in altri casi (come illustra una delle nostre figure) sono isolate le une dalle altre e separate da un tratto di micelio sottile o appena un poco più grosso della norma. In mezzo all'infuso di farina di mais agarizzato si hanno quelle cellule che nella fase di migliore differenziazione hanno una certa somiglianza con clamidospore e che avevamo indicate come ipnospore.

Secondo LANGERON & TALICE queste spore particolari sarebbero state viste e figurate da vari autori e sono nel vero nell'asserire che non si tratta di vere clamidospore. (1)

E'impossibile non paragonare questi due aspetti da un lato alle artrospore ingrossate, opache e giallognole da noi descritte in precedenza per *Geotrichum* (e che sono gli organi di moltiplicazione

---

[1]) LANGERON & TALICE (1932) partendo dal lavoro di REDAELLI & CIFERRI (1929) hanno ristudiato il problema dei *Geotrichum*. Il loro lavoro e scarsamente interessante ai nostri fini poichè tende sopratutto a differenziare i *Geotrichum* dai generi di lieviti con blastospore. Secondo questi AA. *Geotrichum* sarebbe caratterizzato da colonie spesse e membranose, che formano veli sui mezzi liquidi, ma che sono cremose su mezzi solidi per l'abbondanza di coremi. Dalla loro figura emerge che essi indicano come coremi gli aggregati cordoniformi di ife sterili. Questi aggregati sono dunque diversi dai coremi di ife fertili di *Coremiella* che essi non sembrano aver osservato. Anche la loro descrizione delle artrospore lascia ancora dei dubbi, tanto più che dichiarano (pag 73) che assieme alle artrospore si troverebbero anche delle blastospore. E' difficile dunque dire se una parte almeno delle loro culture era mista tra funghi artrosporei e lieviti, o avevano a che fare con qualche Trichosporonacea, famiglia caratterizzata appunto dalla presenza simultanea di artrospore e blastospore e alla quale dovrebbero ascriversi per lo meno alcune delle specie da loro indicate come appartenenti al genere da loro indicato come *Geotrichoides*. Il gen. *Geotrichoides* non è stato mai accettato e sarebbe difficile dirne il significato poichè secondo gli AA. suddetti possiederebbe uno pseudo-micelio, delle blastospore verticillate, delle ,,blastospore-artrospore'' e dei ,,conidi''.

vegetativa del fungo) e dall'altro alle clamidospore apicali e seriate caratteristiche sopratutto di *Candida albicans*. Anche in *Coremiella* come in *Geotrichum* si hanno artrospore clamidospoioidi fertili e artrospore usuali jaline non fertili con la differenza che in *Coremiella* ad un più accentuato aspetto clamidosporioideo delle artrospore fa riscontro una minore differenza tra artrospore sterili e artrospore fertili a tunica ingrossata. Oltre ciò, naturalmente, desideriamo porre l'accento sulle forme a ramificazione penicillata apicale che evolveranno ad aspetti coremioidi di cui si è detto in precedenza.

DISCUSSIONE DEI RISULTATI E TASSONOMIA DEI FUNGHI

Nella contribuzione precendente (CARETTA, 1959) abbiamo fatto cenno al problema sistematico e alla definizione del genere *Coremiella* che HUGHES & TUBAKI pongono nella sezione VII del quadro sistematico degli Ifomiceti (Moniliales). Il genere *Coremiella* fu pubblicato da BUBÁK & KRIEGER (in BUBÁK, 1912) con la seguente diagnosi: "Ife bianche oppure debolmente verdognole, spesso dicotomicamente settate, raffiguranti un coremio con teste inabissate. Le catene dei conidi divise in due dicotomicamente, una fino a due volte. I conidi sono riuniti a forma di catena all'inizio cubici, poi più o meno arrotondati alla superficie".

La specie tipo, *C. cystopoides* BUBÁK et KRIEGER è così caratterizzalsta: "Coremi alti fino a 1/2 mm e larghi fino a 1/5 mm bianchi e debolmente verdognoli. La base bianca, più tardi bruna è larga fino a 8 μ e verso l'alto, nella parte giovane che si continua nella piccola testa larga da 4—5μ; i capitoli sono semisferici; le ife del peduncolo sono spesse volte suddivise dicotomicamente più o meno compatte e intrecciate. Le catene dei conidi sono lunghe, settate, una o due volte dicotomicamente divise, più tardi (come in *Cystopus*) arrotondate più o meno sulla superficie, da 5,5 a 9,5μ in sezione trasversale pareti abbastanza spesse, jaline, biforcantisi in una o due file". Secondo PIRNA si trovano nei gambi, sulla corolla e sul calice morti del *Lythrum salicaria* (1907 sec. KRIEGER).

BUBÁK dice essere un fungo molto interessante che, a causa dei coremi da esso sviluppati, egli situa tra le Stílbaceae jaline e amerospore. HUGHES pubblicò una seconda specie basata sullo *Stysanus ulmariae* McWEENEY (1895) (1) che MASON (in HUGHES, 1953) ha trasportato

---

1) L'isolamento del fungo venne fatto casualmente da McWEENEY nel 1895 in un tratto della palude di Braganstown, Contea di Louth, su *Spiraea ulmariae*. Sulle piante erano visibili piccole macchie puntulate bianco-grigiastre sparse sul fusto, sulle foglie e sui piccioli secchi e grinzosi. Queste piccole macchie puntulate non più alte di 3/4 di mm. erano ben divise tra loro senza tendenza ad aggregarsi e consistevano di una testa glabrosa color grigio argento e di un breve gambo scuro d'aspetto simile a quello di un mixomicete nel suo stato sporifero. Al microscopio erano distintamente visibili una testa e un peduncolo composto da ife parallele o filamenti. Il capitolo è pure composto da filamenti che si irradiano dal peduncolo dando un aspetto capitato dovuto al fatto che la porzione terminale di ciascuna ifa o filamento, è divisa da spessi setti formando una fila di cellule cuboidea di cui quelle terminali

al gen. *Coremiella* e che è così caratterizzata: ,,Conidiofori in ciuffi, ognuno dei quali cresce apicalmente e ramifica irregolarmente o talvolta dicotomicamente divenendo arborescente verso l'apice. I setti trasversali si formano in successione basipeta sui rami superiori, e cellule vicine o alterne, sviluppano pareti più spesse, secondo un processo che ha pure luogo basipetamente. Le cellule intermedie perdono il loro contenuto interamente e le pareti laterali rimangono sottili collassano all'indentro ed infine si rompono lasciando liberi i conidi oblunghi o cubici. Il conidio si forma dentro la parete esterna dell'ifa originale e la caratteristica papilla e il minuto poro della parte terminale della parete sono chiaramente visibili. I conidi che si sono isolati, mostrano un margine o bordo (,,frill)'' ad ogni estremo, che è il residuo della parete esterna dell'ifa originale appartenente ad una delle cellule disfatte. Solamente i rami terminali dei conidiofori si trasformano in conidi''.

Tubaki accetta la sezione VII quale definita da Hughes con la caratteristica di una produzione di conidi per settazione e scissione di ife semplici o ramose, tali conidi essendo stati chiamati artrospore Ma egli suddivide la sezione in due sottosezioni caratterizzate la prima dalla formazione di artrospore per via esogena, per cui si formano setti trasversali nelle ife aeree, secondo una scissione che interessa quasi simultaneamente la parete esterna e il protoplasma dell'ifa generatrice nella formazione dei setti trasversali. Nella secondo sottosezione il meccanismo è endogeno, in quanto i conidi si sviluppano per via endogena dentro la parete esterna delle ife, essendo visibile una papilla o un riunito poro a ciascuna delle estremità dei conidi. Quando il conidio è libero il residuo dell'ifa madre sarebbe visibile come un bordo (frill). Nella prima sottosezione egli include *Geotrichum* (1) e di altri generi che qui non c'interessano e nella seconda sottosezione soltanto *Coremiella*, aggiungendo però

---

presentano angoli arrotondati così da divenire subglobose. Il peduncolo è costituito da ife prallele brune con numerose settazioni: le ife sono lassamente compatte e intrecciate ed alle estremità si biforcano formando una solida massa di rami di cui il capitolo è composto. I rami terminali sono molto piu pallidi in colore di quelli da cui originano e sono divisi da spessi setti in cellule cubiche o a forma di barile. Secondo McWeeney queste cellule successivamente libere avrebbero la funzione di spore e pertanto capaci di germinare nuovamente. Mc Massee, al quale era stato inviato l'esemplare lo ha posto nel gen. *Stysanus* e Mc Weeney ha pertanto denominato il fungo *Stysanus ulmaria* e indicando così il habitat su *Spiraea ulmaria* con la seguente descrizione:
,,*Stysanus ulmariae*, McWeeney, nov. sp. Stipite alto da 0,25 a 1 mm, composto da ife brune, settate, lassamente intrecciate, che verso l'alto si dividono dicotomicamente e si divaricano così da formare una testa glabrosa. Rami terminali più chiari divise da setti spessi in catene moniliformi di elementi cubici che si staccano e costituiscono le spore. Spore bruno-chiare, cubiche a forma di barile o subglobose, 5—6 μ di diam. Su *Spiraea ulmaria* morente, Braganstown palude, Co.Louth, Irlanda, 10 agosto 1895''.

[1] *Oosporoidea* Sumstine (1923) tipificata con *Geotrichum candidum* (sub. *Oidium lactis* Fres.), è naturalmente sinonimo di *Geotrichum*. Il nome del genere fu creato al solo scopo di differenziare i Geotrichi dalle Oospore.

che ulteriori indagini potranno accertare se altri funghi hanno questo stesso tipo di sviluppo.

Per quanto si riferisce al gen. *Allonema* Sydow (1934) genere istituito per il *G. roseum* Grove (1886), Ciferri, Redaelli, Domenici (1943) hanno ammesso che sia da riferire a *Coprotrichum* Bonorden (1831) di cui emendano i caratteri, al qual genere sarebbe pure da riferire per la più parte *Endoconidium* Delacroix (1893). La questione è complessa e comunque esorbita dall' ambito del nostro lavoro, trattadosi di funghi a culture tipo muffe, e con caratteristiche morfologiche affini, ma diverse da quelle dei generi *Geotrichum-Coremiella.*

Confrontando le caratteristiche morfologiche da noi discusse, dei nostri funghi con quelle indicate per *Coremiella* non vi è dubbio che i funghi da noi studiati sono da riferire a questo genere. Per quanto si riferisce alla specie i nostri quattro ceppi sembrano potersi riportare tutti alla *Coremiella ulmariae,* le cui caratteristiche coincidono con quelle da noi osservate per quanto possibile comparare la descrizione data da McWeeney con i dati culturali da noi espressi.

Rimane incerta invece la *C. cystopoides,* troppo brevemente descritta per essere riconoscibile, e che un fortunato futuro isolamento potrá permettere studiare con maggiori dettagli.

Scorrendo le descrizioni delle meno recenti specie che oscillano intorno a questo genere, la nostra attenzione è stata attratta dalle descrizioni di *Oospora cuboidea* Sacc. et Ell. Questa specie è stata descritta da Saccardo (1882) fra i ,,Fungi Boreali-Americani'' con la seguente diagnosi: ,,Candida, effusa, subfarinacea; hyphis subfasciculatis e basi ramosis, filiformibus mox in catenulas conidiorum, 100—120 × 3, abeuntibus; conidiis cuboideis v. globoso-cuboideis, tandem secedentibus, 3 × 2,5—3μ, hyalinis. Hab. in ligno putrescente quercino, Newfield N. J. Nov. 1881 (Ellis, n. 3536)''.

La diagnosi è stata integralmente trascritta da Saccardo stesso (1886) nel Vol. IV della ,,Sylloge Fungorum''.

Tramite il Dr. John A. Stevenson ci fu possibile esaminare l'esemplare autentico di questa specie conservato nel New York Botanical Garden. L'esemplare favoritoci per la cortesia del Curatore dell'Erbario Crittogamico Dr. Clark T. Rogerson consiste in una scaglia di legno disseccata, in gran parte ricoperta da un deposito superficiale gessoso e spesso, di colore bianco sporco che consta interamente delle fruttificazioni del fungo. L'esemplare,eccellentemente conservato, è segnato con il n. 1431b ma con riferimento a Saccardo col n. 3536, e la teca è marcata con il timbro della collezione Ellis. L'etichetta, manoscritta, ha l'indicazione di *Oospora cuboidea* S. & E. e reca le stesse indicazioni di substrato, località e data riferite nella descrizione.

Al microscopio a contrasto di fase appare lussureggiantemente futtificato ed è formato da ife ialine a ramificazione apicale semplice, biforcate, o complesse, nelle cui ife si differenziano artrospore variabili da cuboidee ad arrotondate, in serie lineari anche lunghe, interrotte dai soliti tratti di micelio vacuo e trasparente. Le ife settate sono composte da tratti di micelio vacuo e trasparente e da artrospore opache di 11,2 × 3,2μ. I rami laterali sono formati da artrospore opache seriate composte da 30—50 elementi. Le artrospore componenti i rami laterali sono rettangolari e ad angoli retti nella porzione basale con dimensioni di 4,8μ di lunghezza × 2,4μ di larghezza; nella porzione mediale del ramo assumono la forma cuboidea, μ 2,4 × 2,4 mentre nella porzione terminale presentano ancora aspetto rettangolare, ma con μ 2,4 di larghezza e μ 1,6 di altezza. La settazione è bene evidente e rettilinea. Nel preparato sono presenti, isolate, pure numerose cellule opache rotonde del diam. di μ 2,4 non gemmanti, riscontrabili talora in alcuni frammenti di micelio, intercalati a tratti di micelio vacuo e trasparente (1).

Come è dimostrato anche dalle nostre fotografie, la morfologia di *O. cuboidea* ripete in ogni particolare la morfologia dei ceppi da noi studiati. Anche le dimensioni dei singoli elementi sono le stesse e le settazioni sembrano procedere ugualmente per via centripeta partendo dai rami superiori.

Non si hanno però le forme arbuscolari così ben definite come nelle nostre culture su lamina portaoggetti col metodo RIVALIER-SEYDEL, ma se ne ha soltanto un accenno laddove la vegetazione è rarefatta, il micelio tendendo a disporsi in aggregati reptanti vagamente penicillioidi.

Evidentemente dunque *Coremiella cystopoides*, *C. ulmariae* e *O. cuboidea* sono da riferire allo stesso genere, poichè la modalità di formazione delle artrospore, la loro forma e aggregazione, l'orientamento delle settazioni e la disposizione del micelio sono le stesse. Le sole differenze si hanno nel grado di aggregazione delle fruttificazioni coremioidi. Nella specie tipo sono indicati dei coremi alti sino a 500μ e della larghezza sino di 200μ, e la figure che ne dà BUBÁK è rappresentativa di una Stilbacea mentre la modalità di settazione trasversale del micelio, l'apice tipicamente biforcato in cui ha sede, l'andamento centripeto e la morfologia delle singole artrospore ripete quanto da noi osservato e quanto è descritto o figurato da HUGHES. Nei nostri esemplari gli aggregati coremioidi sono più modesti e, date le modalità della cultura, si sviluppano su due assi ortogonali anzichè su tre. Sono quindi forme rudimentalmente coremioidi, ma infine nettamente arbuscolari o dendroidi. Nell'esemplare autentico di ELLIS si hanno soltanto dei più moderati accenni ad aggregazione di questo tipo sommerso dalla massa di artrospore stratificate al disopra.

Riteniamo quindi che l'aggregazione delle ife fruttifere, cosi evidenti nell'esemplare studiato da BUBÁK da indurlo a porre il genere nella famiglia delle Stilbacee, si attenui in altre specie o ceppi, probabilmente in rapporto a due fattori: uno intrinseco concernente

---

[1]) Un tentativo di cultura dal materiale autentico di SACCARDO ed ELLIS è, naturalmente, andato a vuoto.

la innata tendenza a questo tipo di aggregazioni, ed uno estrinseco in relazione a condizioni dell'ambiente di vita, e più probabilmente a condizioni nutrizionali del substrato.

Che queste ultime condizioni siano fondamentali lo dimostra il fatto che gli aggregati coremioidi si sono formati, nelle nostre culture, su terreni poveri quale l'agar all'infuso di farina di mais a su culture in lamelle di vetro ostili allo sviluppo di qualsiasi fungo per la sottigliezza dell'agar nutritivo se non pure per la difficoltà di mantenerlo in buono stato di idratazione.

Probabilmente non a caso la *Coremiella cystopoides* è stata trovata su una pianta succulenta e carnosa, alofita (quindi ricca di sali) quale il *Lythrum salicaria* ed è questa specie che ha le fruttificazioni aggregate più sviluppate. La *Oospora cuboidea* è invece stata trovata su legno di quercia, substrato di vita povero ed arido, ed è la specie che ha gli elementi di fruttificazione più disgiunti. I nostri ceppi hanno un comportamento intermediario tra i due estremi suddetti.

Ci si può domandare perchè un micologo della tempra di SACCARDO abbia riportato la specie nordamericana al gen. *Oospora* e non a quello che oggi viene definito *Geotrichum*, ma che prima dello studio di LOUBIÈRE (e, quindi, anche negli ultimi decenni dello scorso secolo) si indicava ancora come *Oidium* sensu FRESENIUS, tipificandolo con *l'Oidium lactis* (1850). Probabilmente fu da ciò indotto per gli aggregati delle ife (nella diagnosi indicate come "subfasciculatae") ed alla diformità delle artrospore, entrambi i caratteri non ritrovandosi nell *'Oidium lactis* che SACCARDO aveva studiato in dettaglio nel 1878.

Il riferimento a *Oospora* venne effettuato in virtù di conidi ialini catenulati, in quanto in *Oospora* allora si ponevano tutte le specie con conidi ialini catenulati, come in *Torula* quelli di conidi scuri catenulati. Per marcare le differenze tra le sue specie e quelle usuali di *Oospora* coniò il nome specifico di *cuboidea* che implicitamente era un richiamo a *Oidium-Geotrichum*.

CONCLUSIONI

In conclusione secondo le nostre idee, la diagnosi del genere viene cosi emendata: *Coremiella cuboidea* BUBÀK et KRIEGER emend. CIFERRI et CARETTA.

Artrosporeo a micelio e organi di moltiplicazione vegetativa jalini. Ife fertili, variamente aggregate da densamente tipo stilbaceo a lassamente arbuscolari. Ife sterili moderatamente ramose, irregolarmente ramificate e generalmente forcate agli apici. Settazione progrediente per via centripeta a partire dagli apici con individuazione completa delle artrospore dentro ife miceliche dalle quali si separano per il collasso della tunica materna di cui possono rimanere esigui frammenti all'estremità delle artrospore singole. Artrospore tipicamente cuboidee e a parallelepipedo, spesso ad angoli arrotondati, talvolta a barilotto, ma che possono assumere anche l'aspetto di ipnospore per ingrossamento della tunica e forma ovalata o ellittica sino a sferoidea; tali forme intermediarie tra artrospore e clamidospore sono intercalari, singole o seriate o apicali ed allora una o poche. Attività biochimiche modeste, senza potere fermentativo sugli zuccheri.

Nella sistematica di CIFERRI (1958) è da riferire alla famiglia Geotrichaceae accanto a *Geotrichum* da cui si differenzia per l'assenza di ife fertili aggregate, la diversa modalità di formazione ed andamento della settazione che origina le artrospore.

Provvisoriamente ammettiamo per il genere *Coremiella* due specie:

1) **Coremiella cuboidea** (SACC. et ELL.) CIFERRI et CARETTA, n. comb.
   = *Oospora cuboidea* SACC. et ELL. (1881);
   = *Stysanus ulmatiae* McWEENEY (1895); r
   = *Geotrichum cuboideum* SUMSTINE (1913);
   = *Coremiella ulmariae* (McWEENEY) MASON in HUGHES (1953).
2) *Coremiella cystopoides* BUBÁK et KRIEGER in BUBÁK (1912). Specie incerta da ristudiare.

Nel chiudere questo lavoro è nostro dovere ringraziare il Dr. JOHN A. STEVENSON de The National Fungus Collection, Beltsville, Md., che si è stato focuito tramite e il Dr. CLARK T. ROGERSON, de The New York Botanical Garden per averci cortesemente inviato l'esemplare autentico di ELLIS per lo studio; ma sopratutto il Dr. Prof. R. CIFERRI per l'ausilio prestatoci nello svolgere questa ricerca.

Riassunto

Sono studiati nelle loro caratteristiche morfo-culturali e biologiche quattro ceppi omogenei di *Coremiella* isolati da feci umane. Questo genera è simile a *Geotrichum* ma oltre a cellule miceliche clamidosporo-simili presenta formazione apicali miceliche di tipo coremioide reptante.

L'artrosporulazione è un processo endogeno come in *Geotrichum*, e come in *Geotrichum* si hanno artrospore clamidosporioidi fertili opache e artrospore usuali non fertili e jaline, ma meno differenziate.

I ceppi vengono riferiti a *C. ulmariae* e le caratteristiche vengono paragonate anche con le culture su legno di *Spirea ulmaria*.

Il primo esemplare di *Coremiella* è stato pubblicato come *Oospora cuboidea* SACC. et ELL. come è stato accertato studiando l'esemplare autentico di ELLIS.

In conclusione il genere *Coremiella* BUB.et KRIEG. è stato emendato ed è stata creata la combinazione *C. cuboidea* (SACC. et ELL.).

Summary

Four homogeneous strains of *Coremiella* from human feces were studied from morpho-cultural and biological standpoint. This genus is like *Geotrichum*, but in addition to some mycelial, chlamydospore-like cells, coremioid mycelial, reptant strands are formed.

The arthrosporulation is an endogeneous process, and, like *Geotrichum*, there are usual hyaline and duller chlamydosporioid, fertile arthrospores, but less differentiated.

The strains are referred to *C. ulmariae*, and the characteristics were compared also on timber and woody European Meadowsweet cultures. But the first binomial of *Coremiella* has been published as *Oospora cuboidea* Ell. et Sacc., as ascertained by the study of the authentic Ellis specimen.

In conclusion, the genus *Coremiella* Bub.et Krieg. has been emended, and the combination *C. cuboidea* (Sacc. et Ell.) established.

Bibliografia

Bubák, F. (1912) *Ein Beitrag zur Pilzflora von Sachsen.* Ann. Mycol. Notitiam Sci. Mycol. Univ., **10,** 52.

Caretta, G. (1959) *Caratteristiche morfo-biologiche di ceppi fungini del genere* Geotrichum *isolati da feci umane.* Mycopath. et Mycol. Appl. **XI,** 3, 217—237.

Carmichael, J. W., (1954) *A taxonomic study of the genus* Geotrichum. Thesis, Harvard University.

—, (1955) *Lacto-fuchsin; a new medium for mounting fungi.* Mycologia, **47,** 611.

—, (1957) Geotrichum candidum. Mycologia, **49,** 820—830.

Ciferri, R. & Redaelli, P. (1929) *Studies on the Torulopsidaceae.* Ann. Mycol. **27,** 243—295.

—, (1958) *Sistematica, riconoscimento e frequenza delle Candidae.* Giorn. Malat. Infett. Parassit., **1,** (1).

—, (1958) Manuale di Micologia Medica. Tomo I. Pavia.

Delacroix, M. G. (1893) *Espèces nouvelles observées au laboratoire de Pathologie végétale.* Bull. Soc. Mycol. France, **9,** 184.

Goidanich, G. (1935) *Miceti bolognesi.* Malpighia, **34,** (1), 41.

Grove, B. A. (1886) *New or noteworthy Fungi, III.* J. Bot. **24,** 138.

Hughes, S. J. (1953) *Conidiophores, conidia and classification.* Canad. J. Bot., **31,** 577—659.

Langeron, M. & Talice, R. V. (1932) *Nouvelles méthodes d'étude et essai de classification des champignons lévuriformes.* Ann. Paras., **10,** 1 —80.

Link, H. F., (1809) *Observationes in ordines plantarum naturales.* Berlin. Magazin, **3,** 3—42.

Loubière, A. (1924) *Recherches sur quelques mucedinées caseicocoles.* Thesis, Paris.

Lodder, J. & Kreger van Rij, N. J. W. (1952) *The yeasts, a taxonomic study.* Amsterdam.

McWeeney, M. A. (1895) *A new Irish Fungus.* Irish Naturalist, **4,** 274.

Redaelli, P., Ciferri, R. & Dominici, F. (1943) *La microflora del cadavere umano.* Atti Ist. Bot. Lab. Critt. Univ. Pavia serie 5, **3** (2), 123—125.

Saccardo, P. A., (1882) Michelia Commentarium Mycologicum, **2,** 576.

—, (1886) Sylloge Fungorum, **4,** 13.

Sumstine Ross, D. (1913) *Studies in North American Hyphomycetes.* II. Mycologia, **5,** (2), 45.

Tubaki, K. (1958) *Studies on the japanese Hyphomycetes.* V. *Leaf and Stem Group with a discussion of the classification of Hyphomycetes and their perfect stages.* Hattori Bot. Lab., n. **20,** 142—237.

ABOUT THE "PRECIS DE MYCOLOGIE" BY THE LATE M. LANGERON

R. Ciferri

Centro di Micologia Medica dell 'Universita di Pavia, Italia.

> "We owe respect to the living; we owe the dead nothing but the truth."
>
> Voltaire

(30.VII.1959)

With some delay I was able to read two answers to a review (*Mycopathol. et Mycol. appl.* vol. VII, pag. 214, 1954) to the second edition of the *Précis de Mycologie* by the late M. Langeron. Unfortunately, these answers given by two distinguished scholars not specialized in medical mycology, are rather general, and do not refer to the specific arguments, which alone could have been of some value against our statements.

This is the reason for our being obliged to relate a few precise elements on the onesidedness, or better on the partiality of Langeron, who has systematically tried to ignore the work of Italian medical mycology in general, and that of the late Prof. Redaelli and mine in particular.

But since a complete rectification would be too long, we shall devote our attention only to the principal points. We shall ask you to forgive us, if we shall have to quote ourselves repeatedly [1]).

Regarding the epizootic lymphangitis of Solipeds, Langeron mentions only a summary (L., pag. 606), instead of the complete work of 1934 (R. & C., pag. 447). He further gives tribute to Bullen for the consideration of the relation between human histoplasmosis and epizootic lymphangitis, but the latter, in 1949, did nothing else but confirm what Ciferri & Redaelli had written sixteen years before on the matter.

In discussing the diseases caused by yeasts, Langeron does not refer, even in the bibliography, to the work of Ciferri, Redaelli

---

[1]) For brevity Langeron (L.) is quoted from the ,,Précis de Mycologie", II ed. (Paris 1952) and Redaelli & Ciferri (R. & C.) are quoted from the manual "The fungus granulomata of man in tropical and subtropical regions", pags. 698, Florence (S.E.S.) 1942).

& Cavallero (1938), in which they established the long and complex synonymy, to-day generally accepted, of the species actually called *Candida albicans* (R. & C., page 401—403), and which Langeron (with Guerra), in due course, copied entirely, naturally without mentioning the Italian authors.

Furthermore, Langeron has completely ignored all our series of works on the asporogenic yeasts (Ciferri, 1925; 1930; Ciferri & Redaelli 1925; 1929; 1935; 1939; etc.) including those which paved the way to the modern systematic classification.

In relation to Gilchrist's disease, Langeron reports the synonymous terms which Ciferri & Redaelli established in 1935 (L., pag 476; R. & C., page 306), not mentioning, however, any of their works (cfr. Ciferri & Redaelli, 1935), not even in the bibliography. Furthermore, Langeron had previously [1]) intentionally misunderstood the reasons (of pure nomenclatory priority) which made us substitute the generic name of *Gilchristia* with that (not valid, late homonym) of *Blastomyces* Gilchrist et Stokes, 1898 (not Costantin et Rolland, 1888).

With reference to the so called South American blastomycosis, Langeron twice quotes the works of Redaelli & Ciferri, without ever referring to it in the bibliography, nor ever mentioning their fundamental monograph of 1937.

What causes us strongly to doubt the good faith of Langeron in the matter, is the very curious bibliographical citation of Ciferri & Redaelli's (1936) monograph on the *Coccidioides immitis* and in the coccidioidal granuloma. This work appears in the bibliographical list (L., page 519) under the names of Hartman & Schoo, whose work is dated 1912, although the one by Ciferri & Redaelli was written twenty-four years later. However, aside from the very strange and incorrect way in which the bibliography is made, the determination is evident to obliterate the names of Ciferri and Redaelli. Naturally without mentioning the source, Langeron reports (L., page 510) on all the synonymous terms of the genus and species of the *Coccidioides immitis* (R. & C., page 219—220) established as stated by Ciferri and Redaelli.

We think that this argumentation of ours is sufficient, and it seems to us not useful to answer the arguments, very general indeed, of Blank and Ansell, who never get to the basis of the discussion.

Langeron has had the great merit of studying the human mycoses and the fungi which produce them when very few did investigate the matter, and has done it very often with original, even if not always acceptable, concepts. However, he has had the fault of not wanting ever to recognize the work of those who were not his followers or friends, or of those who did not accept his ideas, although very often he has appropriated the work of others. A proof of what stated above is given also by a French mycologist, and one of the more distinguished one, Dr. GRIGORAKI, who in 1956 wrote: "M. LANGERON who . . . dominated the medical mycology in France, has related my results . . . without ever citing my works". (Mycopath. et Mycol. Appl., Vol. VII, page 307. 1956).

---

[1]) Suppl. Bull. Trim. Soc. Mycol. France, vol. LXV, page 39—40. 1949.

Scholars in good faith will pass their judgement on this matter.

CIFERRI, R. (1925) *Studi sulle Torulopsidaceae. Sui nomi generici Torula, Eutorula, Torulopsis, Cryptococcus e sul nome di gruppo Torulaceae.* Atti Ist. Bot. Univ. Pavia, ser. III, **2**, 129—146.

—, (1930) *Contribuzioni alla sistematica delle Torulopsidaceae.* Arch. Protistenk., **71**, 407—452.

— & REDAELLI, R. (1925) *Studi sulle Torulopsidaceae. Monografia delle Torulopsidaceae a pigmento rosso.* Atti Ist. Bot. Univ. Pavia, serie III, **2**, 147—303.

—, — (1929) *Studies on the Torulopsidaceae. Tentative regarding a diagnostic procedure for species determination.* Zbl. Bakter. Parasit., II Abt., **88**, 40—45.

—, — (1935) *Contributo alla sistematica delle Torulopsidaceae.* Arch. Microbiol., **5**, 1, 9—72.

—, — (1935) *Prima contribuzione allo studio delle cosidette blastomicosi americane. Gli* Endomyces *del gruppo* dermatitidis-capsulatus *agenti della dermatite verrucosa micosica di Gilchrist.* Atti Ist. Bot. Univ. Pavia, serie IV, **6**, 55—105.

—, — (1936) *Morfologia biologica e posizione sistematica di* Coccidioides immitis *Stiles e delle sue varietà con notizie sul granuloma coccidioides* R. Acc. d' Italia, Mem. Cl. Sci. Fis. Mat. e Nat., **VI**, 13, 1.

—, — e CAVALLERO, C. (1938) *L'* Oidium albicans *Robin* Mycotorula albicans (*Rob.*) *Lang. et Tal.*, 1932. *Studio critico e sperimentale.* Mycopath., **1**, 115—161.

—, — (1939) Mycotorula *vs.* Candida: *a plea.* Mycopathol., **2**, 73—74.

—, — (1934) *Sulla posizione sistematica dell'agente patogeno del farcino equino crittococcico.* Boll. Inst. Sierot. Milanese, fasc. 10, pag. 1—8.

—, — (1929) *Studies on the Torulopsidaceae. A trial general systematic classification on the asporigenous ferments.* Ann. Mycol., **27**, 243—295.

—, — (1937) *Morfologia, biologia e posizione sistematica di* Paracoccidioides brasiliensis (*Splendore*) *Almeida* (*Fr. Paracoccidioidaceae*) *con notizie sul granuloma paracoccidioide.* R. Acc. d'Italia Mem. Cl. Sci. Fis. Mat. e Nat., **8**, (12), 557—611.

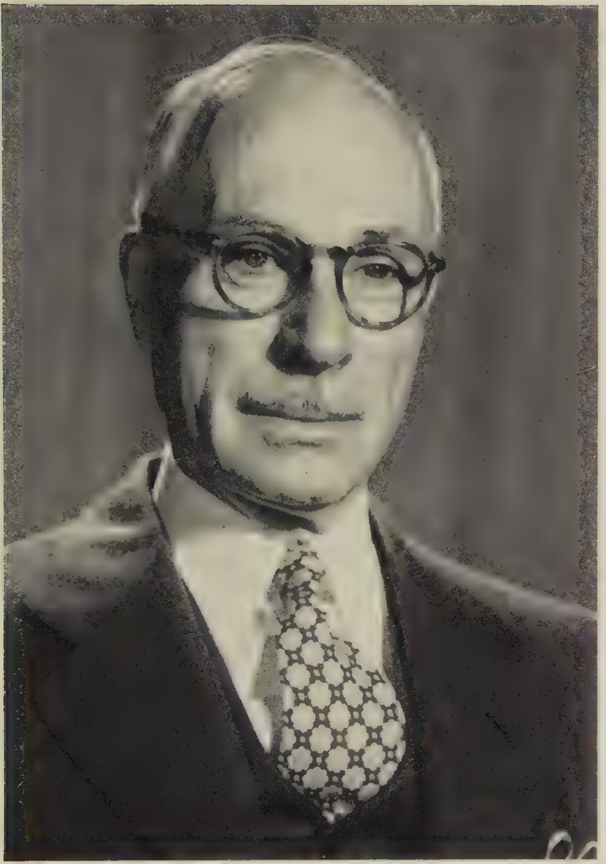
—, — (1931) *Nuovi reperti di* Trichosporon *ed osservazioni intorno a questo genere.* Mycopath., **3**, 203—217.

Here we find also *Cryptococcus neoformans* and *Glenosporella*. Under Dermatophyta the author discusses *Epidermophyton*, *Trichophyton*, *Microsporon* and *Malassezia*. The Actinomycetales close the volume.

In the description of the individual pathogenic fungus he considers its systematic position, clinical and microscopic picture in material and in culture, immunology, therapy and animal experiment. Every chapter closes with a representative selection of literature from the whole world literature. It is particularly useful that the author makes the Slavic language literature accessible to research workers of Latin and Germanic languages.

The text is clear, concise, excellently written. The illustrative material is well selected, most useful. Even though some research worker would disagree with the taxonomic ideas of the author, this certainly does not distract from the high value of his work. The production of the work is highly satisfactory. This work ranks high in the German language mycological literature and gives an excellent picture of the great erudition of its author. This book can be highly recommended to mycologists on any level of their studies.

Tibor Benedek



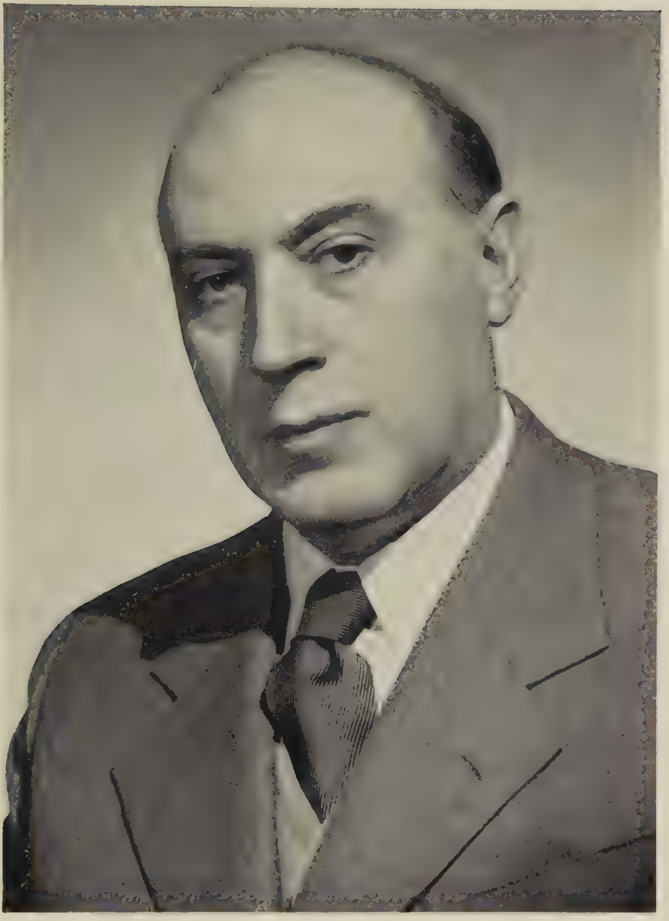
FRANK D. KERN

Botanical Institute

The Pennsylvania State University, Philadelphia, Pa., U.S.A.

Gallery of Contemporary Noted Mycologists

69

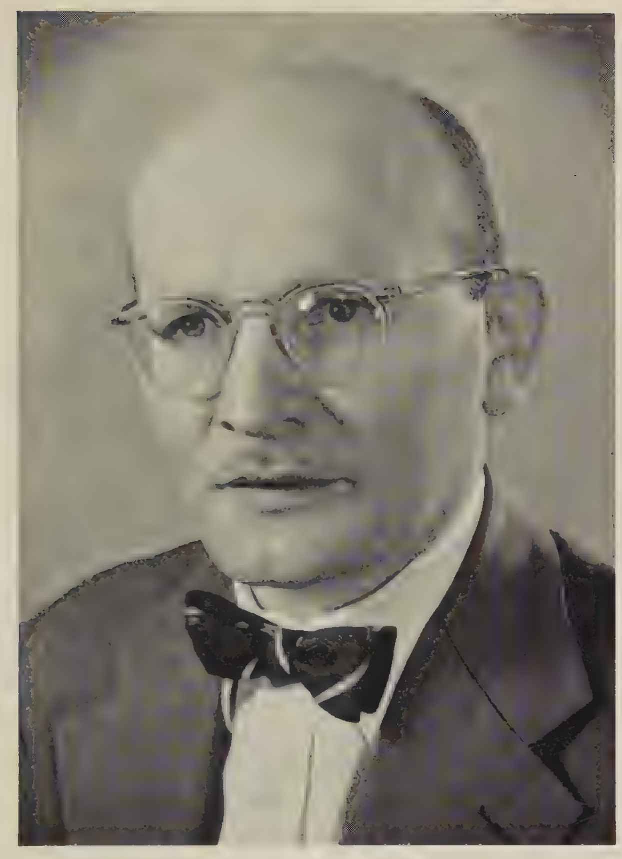


E. FEJÉR

Department of Dermatology
Stephen-Spital, Budapest-Hungary

Gallery of Contemporary Noted Mycologists

70



ALFRED S. SUSSMAN

Department of Botany
University of Michigan, Ann Arbor, Mich., U.S.A

Gallery of Contemporary Noted Mycologists

71

HERNAN MIRANDA

Departamento de Microbiología
Facultad de Medicina, Trujillo, Perú

Gallery of Contemporary Noted Mycologists

72

SENSITIZATION AND ANAPHYLACTIC SHOCK INDUCED IN GUINEA PIGS BY USING FUNGUS EXTRACTS

by

S. Landi & J. D. L. Fitzgerald

(*Department of Botany, University of Toronto and Department of Allergy, Western Hospital, Toronto, Ontario, Canada.*)

(27.VIII.1959)

Introduction

The purpose of this work was to find out whether guinea pigs could be readily sensitized by inoculating them with fungus extracts and see if the anaphylactic shock could be used as a means of species determination.

Material and Method

The organisms for this study are listed together with their sources in the following table.

Table I.

Stock	Organism	Source
12435	*Aspergillus flavus*	Dr. R. F. Cain, University of Toronto
12484	,, *niger*	,, ,,
12261	,, *rugolosus*	,, ,,
12428	,, *niveus*	,, ,,
12440	,, *ustus*	,, ,,
32244	,, *amstelodami*	,, ,,
W.P.C.38	,, *clavatus*	,, ,,
	,, *quadricinctus*	,, ,,
	,, *nidulans*	,, ,,
	,, *fumigatus*	Mr. J. Fischer, Ont. Dept. of Public Health, Toronto.

The aspergilli were grown in Roux bottles containing Sabouraud maltose broth, Difco.

Proteins were extracted by using Coca extracting fluid.

Millers containing large beads and a rolling shaker, kept in a cool room at 7° C, were used to crush the mycelial mats.

Fifty-five guinea pigs varying in weight from 250 to 300 grams were used.

Five cc syringes and gauge # 24 needles were used.

Two batches of aspergilli were grown for a period of 60 days at room temperature in Roux bottles, each bottle contained 230 cc of Sabouraud maltose broth. Each batch comprised 50 Roux bottles and 5 bottles per batch were assigned to each species of aspergilli. When the 60 day period of incubation was reached, groups of 5 mats were removed from the bottles and placed in sterile Erlenmeyer flasks containing 350 cc of sterile distilled water. The flasks were moderately shaken for 5 minutes and the water was discarded. The mats were thoroughly washed twice more to be sure that no medium was present on which the fungi had grown. The washed mats were then placed into separate sterile millers, each containing 16 large beads and 125 cc of Coca's extracting fluid, and were placed on a rolling shaker for 3 days. The crushed mats and extracting fluid were centrifuged at 3000 r.p.m. at 15° C, for a period of 20 minutes. The sediments were discarded and the supernatants dialyzed and Seitzed. The total nitrogen per cc of these extracts was determined by the micro-Kjeldahl method. The extracts were singly tested for sterility by adding 0.5 cc of each extract to 30 cc of liquified nutrient agar medium and by pouring it into a 10 cm in diameter petri plate. For each extract one plate was incubated at room temperature and one at 37° C. The extracts were then dispensed in 10 cc vials, sealed with rubber caps and Cel-O-Seal bands, and placed in a refrigerator. For each species of aspergilli some extract was freeze dried and the residue suspended again in a volume of sterile distilled water equal to one third of its initial volume; thus the shocking dose contained the same amount of total nitrogen as the sensitizing dose in a smaller volume of fluid. These newly concentrated extracts were Seitzed, tested for sterility and dispensed in vials as above described.

Fifty-five guinea pigs were separated into 10 groups, each group comprising one guinea pig less than the previous group. The first group had 10 guinea pigs and the last group 1 guinea pig. In this manner we were able to test the specificity of all our extracts with the minimum number of guinea pigs. The guinea pigs were sensitized by injecting subcutaneously 1 mg of total nitrogen into the inner part of the thigh and after a lapse of time varying from 35 to 42 days, a shock dose containing 1 mg of total nitrogen was injected intracardially.

RESULTS

TABLE II.

Total nitrogen as determined by the micro-Kjeldahl method.

Name of the organism	Symbol	Total nitrogen mg./cc	Sensitizing dose in cc = 1 mg. total nitrogen	Shocking dose in cc = 1 mg. total nitrogen *
Aspergillus niger	A	.35440	2.82	.94
Aspergillus flavus	B	.61346	1.63	.54
Aspergillus fumigatus	C	.56704	1.76	.58
Aspergillus rugolosus	D	.86544	1.15	.38
Aspergillus nidulans	E	.45221	2.21	.73
Aspergillus niveus	F	.22823	4.38	1.46
Aspergillus quadricinctus	G	.38098	2.62	.87
Aspergillus amstelodami	H	.18180	5.5	1.83
Aspergillus ustus	I	.30939	3.23	1.08
Aspergillus clavatus	J	.37247	2.92	.97

* Three times more concentrated than the sensitizing dose.

TABLE III.

Anaphylactic shock induced in guinea pigs.
Aspergillus niger.

No. of g.p. sensitized	Sensitizing period in days	Extract used as sensitizing dose	Extract used as shock dose	Results
10	39	A	A	+++
		A	B	++
		A	C	++
		A	D	—
		A	E	—
		A	F	—
		A	G	—
		A	H	—
		A	I	—
		A	J	+

(—) No anaphylactic shock.
(+) Mild anaphylactic shock.
(++) Strong anaphylactic shock. Very frequent convulsions with dyspnea, dropping on its side and recovering.
(+++) Death.

TABLE IV.

Aspergillus flavus

No. of g.p. sensitized	Sensitizing period in days	Extract used as sensitizing dose	Extract used as shock dose	Results
9	42	B	B	++
		B	C	—
		B	D	—
		B	E	—
		B	F	—
		B	G	+
		B	H	—
		B	I	—
		B	J	++

(—) No anaphylactic shock.
(+) Mild anaphylactic shock.
(++) Strong anaphylactic shock. Very frequent convulsions with dyspnea, dropping on its side and recovering.

TABLE V.

Aspergillus fumigatus

No. of g.p. sensitized	Sensitizing period in days	Extract used as sensitizing dose	Extract used as shock dose	Results
8	42	C	C	+++
		C	D	—
		C	E	—
		C	F	—
		C	G	—
		C	H	+
		C	I	—
		C	J	+

(—) No anaphylactic shock.
(+) Mild anaphylactic shock.
(+++) Death.

TABLE VI.

Aspergillus rugolosus

No. of g.p. sensitized	Sensitizing period in days	Extract used as sensitizing dose	Extract used as shock dose	Results
7	39	D	D	+
		D	E	—
		D	F	+
		D	G	—
		D	H	++
		D	I	—
		D	J	—

(—) No anaphylactic shock.
(+) Mild anaphylactic shock.
(++) Strong anaphylactic shock. Very frequent convulsions with dyspnea, dropping on its side and recovering.

TABLE VII.

Aspergillus nidulans

No of g.p. sensitized	Sensitizing period in days	Extract used as sensitizing dose	Extract used as shock dose	Results
6	42	E	E	++
		E	F	—
		E	G	+
		E	H	++
		E	I	—
		E	J	—

(—) No anaphylactic shock.
(+) Mild anaphylactic shock.
(++) Strong anaphylactic shock. Very frequent convulsions with dyspnea, dropping on its side and recovering.

TABLE VIII.

Aspergillus niveus

No. of g.p. sensitized	Sensitizing period in days	Extract used as sensitizing dose	Extract used as shock dose	Results
5	35	F	F	+
		F	G	—
		F	H	++
		F	I	—
		F	J	—

(—) No anaphylactic shock.
(+) Mild anaphylactic shock.
(++) Strong anaphylactic shock. Very frequent convulsions with dyspnea, dropping on its side and recovering.

TABLE IX.

Aspergillus quadricinctus

No. of g.p. sensitized	Sensitizing period in days	Extract used as sensitizing dose	Extract used as shock dose	Results
4	35	G	G	+++
		G	H	+
		G	I	—
		G	J	—

(—) No anaphylactic shock.
(+) Mild anaphylactic shock.
(+++) Death.

TABLE X.

Aspergillus amstelodami

No. of g.p. sensitized	Sensitizing period in days	Extract used as sensitizing dose	Extract used as shock dose	Results
3	35	H	H	++
		H	I	—
		H	J	+

(—) No anaphylactic shock.
(+) Mild anaphylactic shock.
(++) Strong anaphylactic shock. Very frequent convulsions with dyspnea, dropping on its side and recovering.

TABLE XI.

Aspergillus ustus

No. of g.p. sensitized	Sensitizing period in days	Extract used as sensitizing dose	Extract used as shock dose	Results
2	36	I	I	+
		I	J	++

(+) Mild anaphylactic shock.
(++) Strong anaphylactic shock. Very frequent convulsions with dyspnea, dropping on its side and recovering.

TABLE XII.

Aspergillus clavatus

No. of g.p. sensitized	Sensitizing period in days	Extract used as sensitizing dose	Extract used as shock dose	Result
1	36	J	J	++

(++) Strong anaphylactic shock. Very frequent convulsions with dyspnea, dropping on its side and recovering.

SYNOPTIC TABLE XIII.

Results Shock dose Sensitizing dose	Results Shock dose Sensitizing dose	Results Shock dose Sensitizing dose	Results Shock dose Sensitizing dose	Results Shock dose Sensitizing dose	Results Shock dose Sensitizing dose	Results Shock dose Sensitizing dose	Results Shock dose Sensitizing dose	Results Shock dose Sensitizing dose	Result Shock dose Sensitizing dose
A A +									
A B +	B B +								
A C +	B C —	C C +							
A D —	B D —	C D —	D D +						
A E —	B E —	C E —	D E —	E E +					
A F —	B F —	C F —	D F +	E F —	F F +				
A G —	B G +	C G —	D G —	E G +	F G —	G G +			
A H —	B H —	C H +	D H +	E H +	F H +	G H +	H H +		
A I —	B I —	C I —	D I —	E I —	F I —	G I —	H I —	I I +	
A J +	B J +	C J +	D J —	E J —	F J —	G J —	H J +	I J +	J J +

A = *Aspergillus niger*
B = ,, *flavus*
C = ,, *fumigatus*
D = ,, *rugolosus*
E = ,, *nidulans*
F = ,, *niveus*
G = ,, *quadricinctus*
H = ,, *amstelodami*
I = ,, *ustus*
J = ,, *clavatus*
+ = anaphylactic shock
— = no anaphylactic shock.

SUMMARY AND CONCLUSION

1. It has been found that guinea pigs can be readily sensitized by the extracts of the following species of aspergilli, namely: *Aspergillus niger* (A), *Aspergillus flavus* (B), *Aspergillus fumigatus* (C), *Aspergillus rugolosus* (D), *Aspergillus nidulans* (E), *Aspergillus niveus* (F), *Aspergillus quadricinctus* (G), *Aspergillus amstelodami* (H), *Aspergillus ustus* (I), *Aspergillus clavatus* (J).

2. The sensitization period varied between 35 to 42 days, although no effort was made to find out if they could have been sensitized in a shorter period of time.

3. Guinea pigs sensitized to the above species of fungi were injected intracardially with a shock dose of the same or different extracts and as shown in Tables III to XIII ,anaphylactic reactions were present in all cases where the extract used as a shock dose was obtained from the same species used for sensitizing the guinea pigs.

4. Cross-reactions were also present, as shown in Tables III to XIII, as follows:

Aspergillus niger (A) cross-reacting with *Aspergillus flavus* (B), *Aspergillus fumigatus* (C), *Aspergillus clavatus* (J).

Aspergillus flavus (B) cross-reacting with *Aspergillus quadricinctus* (G), *Aspergillus clavatus* (J).

Aspergillus fumigatus (C) cross-reacting with *Aspergillus amstelodami* (H), *Aspergillus clavatus* (J).

Aspergillus rugolosus (D) cross-reacting with *Aspergillus niveus* (F), *Aspergillus amstelodami* (H).

Aspergillus nidulans (E) cross-reacting with *Aspergillus quadricinctus* (G), *Aspergillus amstelodami* (H).

Aspergillus niveus (F) cross-reacting with *Aspergillus amstelodami* (H).

Aspergillus quadricinctus (G) cross-reacting with *Aspergillus amstelodami* (H).

Aspergillus amstelodami (H) cross-reacting with *Aspergillus clavatus* (J).

Aspergillus ustus (I) cross reacting with *Aspergillus clavatus* (J).

5. No well defined pattern of cross-reactions could be established and therefore anaphylactic shock cannot be used as a specific reaction for the above species of aspergilli.

THE GENUS BLASTOMYCOIDES AS A MYCOLOGICAL ENTITY

by

MAHMOUD KAMAL MUFTIC

Gaza, Palestine, U.A.R.

(5.X.1959)

The importance of filamentous yeast-like fungi as etiological factors in different dermatothrophic and visceral mycoses was considerably increased especially after the introduction and abuse of the antibiotic therapy, From the clinical point of view it was conveniant to regard all these diseases as a clinical entity. This group of diseases caused by filamentous yeast-like fungi is better known under the name Blastomycoses. From mycological point of view they are very rigorously discernated however and a uniform conclusion about different species as causal factors of blastomycoses was not achieved.

Blastomycoides, as a genus of the Ascomycetes, was described by LINK (1816) with the type-species *Blastomyces luteus*. When VUILLEMIN (1919) found that it was identical with *Aleurisma flavissimum* CHEVALIER 1836, he proposed the abolishment of the genus *Blastomycoides*. This conception unfortunately induced many mistakes in mycological classification, because some species of *Blastomycoides* were designed, according to this theory, as *Aleurisma*, others as *Scopulariopsis* or were put in various mono-generic and mono-specific families. To ameliorate this situation, BOGLIOLO & AROEIRA-NEVES (1952) proposed the inclusion of several species of the old genus *Blastomycoides* into the Aleurismaceae, which will include three of the etiologic factors of blastomycoses (*Bl. dermatitides, Bl. brasilienses* and *Coccidioides immitis*). This classification was, of course, established only on the bases of morphogenetic characteristics of the filamentous phase of their growth, strictly corresponding to VUILLEMIN's Conception.

With the discovery of *Blastomyces cerolytica* (MUFTIC, 1949), the genus *Blastomycoides* (CONSTANTIN & ROLLAND 1889) has to be reconsidered. It is not approved and accepted that the filamentous phase is the only criterium for determination of fungi, especially when they have a yeast phase of growth. Sexual forms of *Blastomycoides* show special characteristics, but these were not considered in the determinations of the species. It is not my intention to say that Vuillemin's conception is incorrect from botanical point of view; however, it is well known that the vegetative forms in the great majority of fungi are very similar and that these can not be the only factor in morphological differentiation between genera and species. For this reason I believe that we can forward a proper determinative judgement about the systematical place of a fungus only after consideration of the whole life cicle (vegetative, yeast and sexual phase). As to *Blastomycoides*, the difficulty was to obtain the yeast phase in vitro as well as perfect sexual forms in the saprophytic phase of their life. Morphological and genetical data concerning several *Blastomycoides* were not available up till now. Several attemps were made to obtain in vitro yeast forms and the sexual phase in different filamentous fungi. It is known that in generally used mycological media they only grow as vegetative mycelium very much resembling Aleurismaceae, which fact has influenced Bogliolo & Aroeira-Neves to admit them to the Aleurismaceae. Meanwhile Mackinnon & Edwards (1950) succeeded in obtaining the interconversion of the filamentous to the yeast phase in *Blastomycoides dermatitidis* and *Bl. brasiliensis* by varying different thermal and physical factors. Titswort & Greenberg (1950) obtained the yeast phase of *Histoplasma capsulatum* in continuity on egg-yolk-potato medium. De Monbreun (1935) introduced a method which can be used for the continous cultivation of the yeast phase of different filamentous fungi. He inoculated mycelial elements into sensitive animals and then reinoculated infested material into protein-rich media such as: rabbitblood agar of Petragnani medium (without malachit green). Incubation was always at 37° C. Nickerson & van Rij (1949) preserved the yeast phase in continuity by addition of different reducing and chelating subtances as cystein, glutathion, sodium thioglycollate and 8-hydroxyquinaline to the media. Rondoni & Barbieri (1950) used BAL[1] for the same purpose. Scherr (1952) later stated that high temperature (above 35°C), alkalinity of the media and abundance of :SH'-radicals support the growth of the yeast phase and inhibit the filamentous progrediance. Conant (1941) observed the yeast phase of *H. capsulatum* in sealed tubes of blood agar, but not in unsealed ones. Bullen (1949) was able to grow the yeast-like phase of the dimorphic pathogenic fungi *H. capsulatum, B. dermatitidis* and *Sporotrichum schenckii* on peptone agar by controlling the carbon dioxide content of the atmosphere above the culture. Macneal & Taylor (1914) observed multiplication of the parasitic form of *Coccidioides immitis* in infected pus when added to ascitic fluid or gelatinised horse serum containing sterile kidney slices. Lack (1938) reported on development of these forms from arthrospores inoculated into Hall tubes containing glucose broth and partially coagulated egg

---

1) BAL = British Anti-Lewisite.

albumin. BAKER & MRAK (1941) observed development of this phase of *Coccidioides immitis* in a variety of solid media when active growth has ceased as the result of the drying of the media. BURKE (1951) reported the appearance of spherules in a partially defined solid medium containing coconut milk. CONANT & VOGEL (1954) observed spherules in cultures transferred to Sabouraud's medium following exposure to Tween 80. LUBARSKY & PLUNKETT (1955) produced parasitic phase growth of *C. immitis* in Tyrodes solution supplemented with chick-embryo extract and serum. CONVERSE (1955) succeeded in obtaining pure parasitic phase growth in vitro using lithium hydroxide and Tamol "N" (sodium salt of a condensed arylsulfonic acid) with complete exclusion of mycelial forms.

By oxidative desamination of tryptophane different fungi form auxin and auxin-like substances which, according to SCHERR (1952) are identical with so-called "elongation factors". These compounds induced elongation forms in the dimorphic yeast-like fungi as well as in pure yeasts and represent the main factors in the formation of the mycelial stage of growth. During aging the concentration of auxin-like substances increases and for that reason elongation forms are observed in older cultures. This phenomenon was described by NADSON & KRASSILNIKOV (1932) and PUNKARI & HENRICI (1933).

ZIKES (1920) observed the formation of elongation forms at low temperatures (9 to 20°C). SCHERR (1952) described the same fact using low pH concentrations. Different metallic ions can influence the formation of elongation forms; NICKERSON & VAN RIJ (1949) observed this phenomenon in cultures containing cobalt salts and MICKLES & JONNES (1940) in cultures with lithium and boron salts. Several organic compounds and especially narcotics have the same action. LEVAN & SANDWALL (1943) discovered a reaction designed as "camphor reaction" or "cf", which is related to the acceleration of elongation formation in different yeasts and yeast-like fungi by camphor and more or less related substances. Over 50 organic compounds were found to have "cf" properties: SEGAL (1939) found them in fusel oil and higher aromatic alcohols, DODGE (1952) in in methylcholantrene, COCKART & JOLLIJ (1939) in dibenzantrene, SCHERR (1952) in other carcinogens, NICKERSON & VAN RIJ (1949) in penicillin and MICKLE & JONNES (1940) in immune rabbit serum.

EXPERIMENTAL

In order to obtain the yeast phase of *Blastomycoides* in vitro in continuity I assumed that elimination of the factors which help the production of auxins and auxin-like substances would be the best method. Inactivation of the enzymes participating in the oxidative desamination of tryptophane was the first aim. For this reason different antioxidative compounds were used. Generally

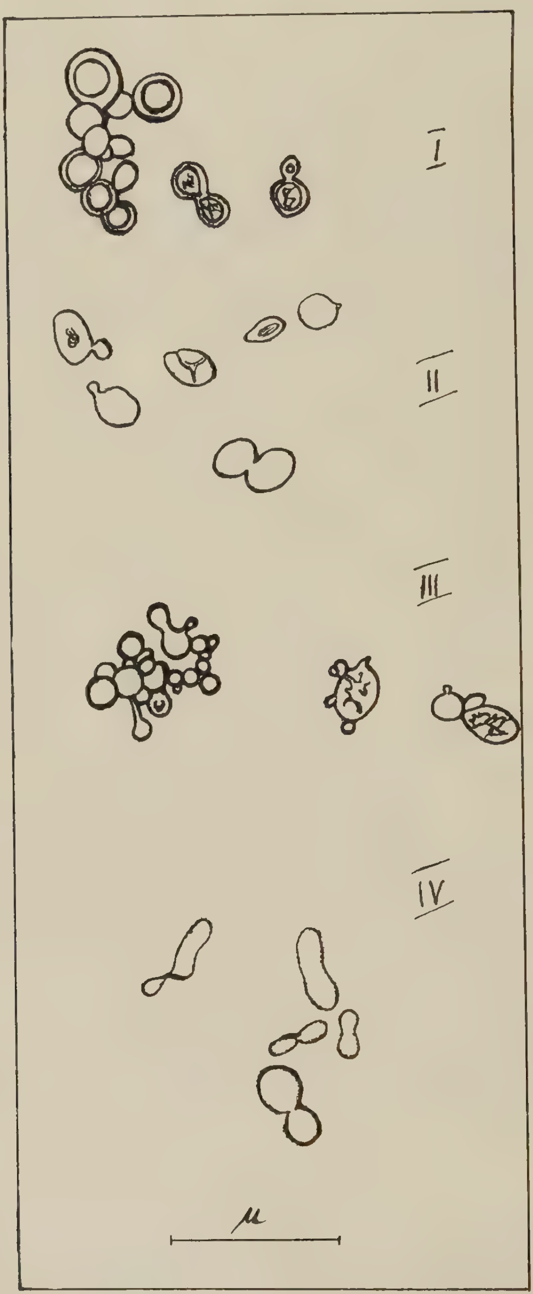


Fig. 1. Camera Lucida drawings of the yeast phases of Blastomycoides

I = *Blastomyces dermatitidis*
II = *Blastomyces immitis*
III = *Blastomyces brasiliensis*
IV = *Blastomyces cerolytica*

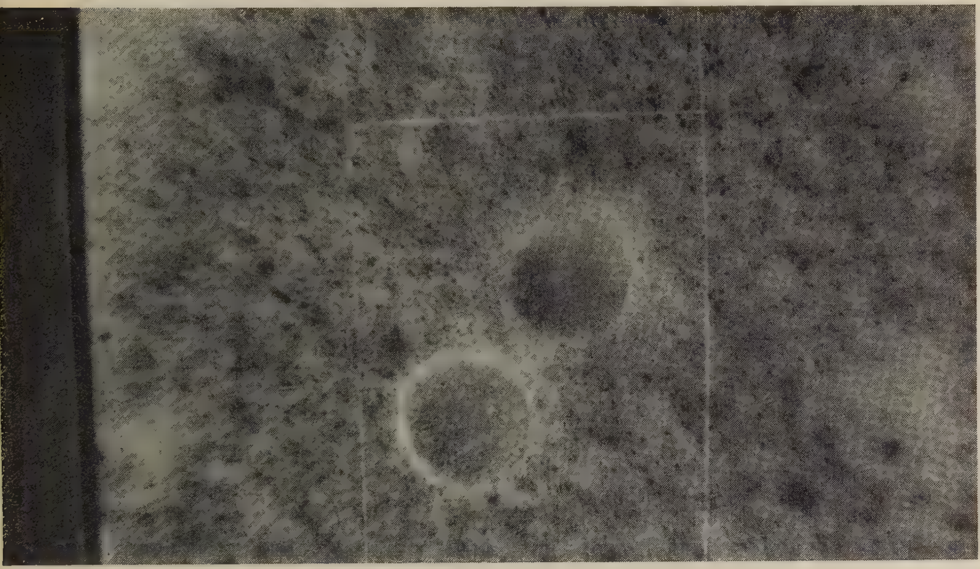


Fig. 2. Asci of *Blastomyces immitis* from arteficial media. 700 ×

these are chelating compounds inactivating the enzymes by removal of the metallic ions responsible for the activation of the enzyme, as cystein, glutathion, sodium thioglycollate, BAL, salicylaldoxime, quinaldic acid, 8-hydroxyquinaldine and 8-hydroxyquinoline. Two other desaminase inhibitors were used, for which the chelating power has not been assured: urea and semicarbazide. As basal medium Wickerhams solution was used. Incubation was at room temperature (38—40° C in subtropical regions) and also in a incubator fixed at 37° C. The influence of the variation of the pH on the relationship filamentous/yeast phase was also studied.

Auxanographic tests (in the yeast phase of *Blastomycoides*) have been done according to the method described by NEGRONI & DAGLIO (1949). The substances to be investigated are put into small glass cylinders (resembling those used for antibiotic tests), already planted in Wickerhams agar plates.

In the morphogenetic study of the yeast phase of *Blastomycoides* a close relationship of cytological elements was found. The yeast forms were investigated at different time intervals after inoculation and special attention was paid to the budding forms. Yeast colonies of *Blastomycoides* can be with difficulty distinguished macroscopically. Only in microscopical preparations are there typical morphological differences which can be a basis for identification. As to the vegetative spores (aleurospores and chlamydospores) of the filamentous phase these are common to a great variety of fungi, often very advanced in evolution and differentiation.

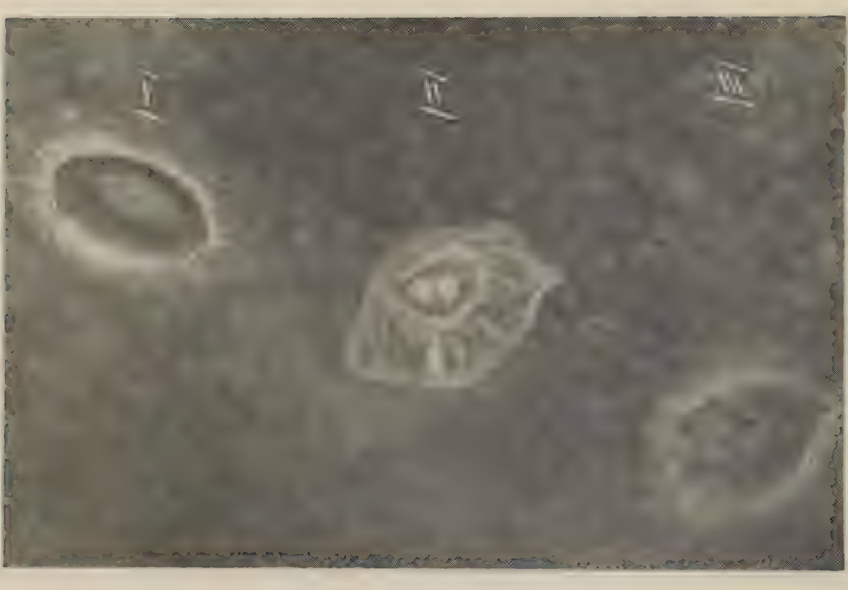


Fig. 3. *Blastomyces immitis.*

I = Ascospores
II = Transformation of nucleus by degradation of nuclear membrane and dissociation of chromosomes. FIALHO silver impregnation. 2400 × (U.V. microscope).

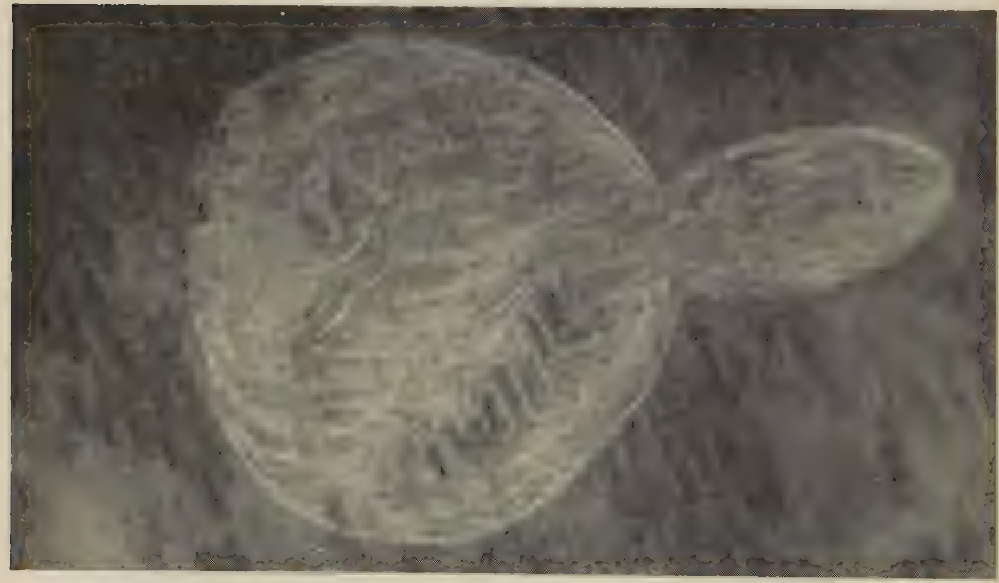


Fig. 4. *Blastomyces immitis.* Yeast cell in budding.

The films of the yeast-forms of *Blastomycoides* are fixed by flame or by osmic acid vapour and then stained by NEGRONI's (1949) method with SudanIII and Gueguens reagent. Other slides are treated by FIALHOS (1947) silver-diethanolamide impregnation. The characteristic cytological elements are mentioned in table III. From the cellular elements observed in the yeast-cells we found simple Gram-positive argentophyllic granulations and acidophyllic spherules. These elements seem to be formed from nucleotides as was established by Feulgen staining. In *Blastomyces dermatitidis* and *Blastomyces cerolytica* only a kind of fine granulation could be observed. In *Blastomyces brasiliensis* and *Coccidioides immitis* voluminous and very acidophyllic spheres were found. Fig. 3 shows this spherule in the yeast-form of *Coccidioides immitis* (or *Blastomyces immitis*), with a clear differentiation between the three stages of formation. The first on the left is an ascospore, the middle one a nucleus under transformation (desaggregation) and the one on the right represents a typical yeast cell. The ultramicroscopic picture as well as the electron-microscope picture (Fig. 4) show the same budding yeast cells of *Blastomyces immitis*.

Whenever the suspicion raised whether ascospores were really spores of fatty droplets or some other particular cellular conglomeration, we have used Monawets (anionic detergents) for the lysis of the cell wall and dispersion of the cellular elements in a conveniant detergent (Monapal), using this suspension as inoculation material to prove whether these elements can grow or not. This was done in view of DE MONBREUN's conception, concerning the sexual forms of *Blastomyces dermatitides*, where true ascospores were interpreted as fatty droplets without proof for this claim. To obtain asci in *Blastomycoides* we used Tween 85-spermin agar. After incubation, the surface of the culture was washed with a 40% glucose solution containing 2% Monapal (non-ionic detergent) and filtered through a Glass filter No. 00 to prevent mycelial particles from passing through. Single spore cultures in hanging drops as well as slide cultures were used with different synthetic or enriched media. As a routine we used Wickerhams solution and Sabouraud agar. Dependent from the presence or absence of the inhibitors of the elongation factors a yeast phase growth or a filamentous phase growth was obtained. In *Blastomyces cerolytica*, as was previously described by MUFTIC (1957), the formation of the dycarion phase was dependent on the presence of the so-called "caryogamy factors", as is the case in other *Blastomycoides*. This factor may be extrinsic as in *Bl. dermatitidis* and *Bl. cerolytica* or intrinsic as observed in *Bl. brasiliensis* and *Bl. immitis*.

RESULTS

The inhibitors of tryptophane oxidase-transaminase were able to inhibit the mycelial growth of *Blastomycoides*. They of course differ in efficacy according to the species and to factors such as pH,

Nitrogen Substances Auxanogram

Substance	*Blastomyces*			
	dermatitidis	*brasiliensis*	*cerolytica*	*immitis*
Potas. nitrate	—	—	—	—
Potas. nitrite	—	—	—	—
Potas. rhodanide	—	—	—	—
Nitrocellulose	—	—	—	—
Ammonium chloride	+ +	+ +	+ +	+
Urea	+	±	+	—
Glycol	+	+	+	—
Peptone	+ +	+	+ +	+ +
Histamine	—	—	—	—
Ethylendiamine	—	—	±	±
Spermine	+	+	+	+

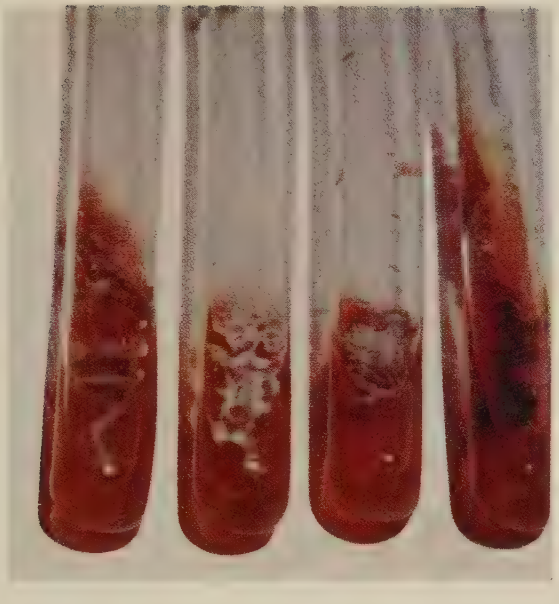
Hydrocarbon and carbohydrate substances Auxanogram

Substance				
Glucose	+ +	+ +	+ +	+ +
Saccharose	+	+	+ +	±
Lactose	+	+	+	+
Galactose	+	+	+	+
Maltose	+ +	+ +	+ +	+ +
Raffinose	+	+	+ +	+
Mannitol	+ +	+ +	+ +	+ +
Dulcit	+	+	+	±
Dextran	+	+	+ +	+
Amylum	±	±	+	±
Glycerol	±	±	±	±
Ethanol	—	—	±	—
Amyl alcohol	—	—	—	—
Sod. acetate	—	—	—	—
Sod. benzoic	—	—	—	—
Oleic acid	—	—	±	—
Paraffine	—	—	+ +	—

Microscopic morphology of yeast forms of *Blastomycoides*

Blastomyces	form	capsule	budding	aspect
dermatitidis	round	thick	monopolar	granules
brasiliensis	ovale	no	multipolar	spherules
cerolytica	elipsoid	thin	monopolar	granules
immitis	ovale	no	monopolar	granules

Plate I.



Represent yeast phase of the Blastomycoides cultivated on Wickerham's modified medium (Semicarbazide; BAL).

I = *Blastomyces dermatitidis*
II = *Blastomyces brasiliensis*
III = *Blastomyces cerolytica*
IV = *Blastomyces immitis*

pH concentration range permitting yeast form growth in *Blastomycoides*

Fungus	pH 1 2 3 4 5 6 7 8 9 10 11 12 13 14
Bl. dermatitidis	+ + + + + + +
Bl. brasiliensis	+ + + + + + + + + + + + + +
Bl. cerolytica	+ + + + + + + + + + + + + +
Bl. immitis	+ + + + + + +

Yeast-phase growth Promoting Factors

Fungus	I	II	III	IV	V	VI	VII	VIII	IX	X
Bl. dermatitidis	++	++	++	++	±	±	+	+	+	++
Bl. brasiliensis	++	++	+	++	—	—	±	+	±	++
Bl. cerolytica	++	++	+	+	+	±	—	— —	±	++
Bl. immitis	—	—	—	±	—	—	—	±	—	+

LEGENDA:
I = cystein
II = gluthation
III = sod. thiogycol.
IV = BAL
V = salicylaldoxime
VI = quinaldic acid
VII = 8-hydroxyquinaldine
VIII = 8-hydroxyquinoline
IX = urea
X = semicarbazide

Surface Active Agents inhibiting mycelial growth

S.A. Agent	*Blastomyces*			
	dermatitidis	*brasiliensis*	*cerolytica*	*immitis*
Monamine AD-100	+	+	+	+
Monamine AA-100	+	+	+	+
Monamine ACO-100	±	±	+	—
Monamid 150-M	—	—	—	—
Monamid 150-CE	—	—	+	+
Monamid 150-ADD	—	±	±	±
Monawet Mo-70	+	++	+++	+
Monawet MM-80	+	++	++++	++
Monamine ALF	+	++	++	+
Monapal T	++	+++	++++	+++
p-p'-Dioctyl-diphenylamine	—	+	++	+
Tween 20	++	++	++	++
Tween 80	+++	++	++	++
Tween 85	++++	+++	++	+++
Arlacel S	+	+	+	+
Brij F	+	++	+	+
Myrj G	+	++	++	++
Span W	++	++	++	++
Triton WR 1339	+++	+++	+++	+++

LEGENDA:
— = no activity
± = trace of inhibition
+ = evident inhibition
++ = strong inhibition
+++ = very strong inhibition
++++ = exclusive yeast forms

The surface active agents in No. 1 to 10 were obtained from MONA Industries, U.S.A., those in No. 11 to 19 from HONEYWELL & STEIN Ltd., London.

All surface active agents are used in 5% concentration in liquid Wickerham's medium without addition of any other agents. After one week incubation the test tubes were centrifuged, the supernatant filtered through Glass filter No. 00 and the filtrate then inseminated in large Petri dishes with Wickerham's agar.

temperature and pretreatment of the inoculum. Cystein was able to maintain yeast-phase growth in all species of *Blastomycoides* except *C. immitis*. 8-hydroxyquinaldine promotes yeast growth in *Bl. dermatitides* and partially in *Bl. brasiliensis*, but has no effect in *Bl. cerolytica* and *C. immitis*. 8-hydroxyquinolin maintains the yeast phase in all *Blastomycoides* except *Bl. cerolytica*. The most effective agent for all species of *Blastomycoides* was semicarbazide, for what reason we use it in all further studies as a standard method. Yeast-like growth needs a limited pH concentration, which sometimes is very narrow. *Bl. dermatitidis* e.g. needs a pH range between 6—8, *Bl. brasiliensis* from 4—8, *Bl. cerolytica* from 3—7, *C. immitis* from 6—8. As far as auxanographic charts are concerned, there is a very close relationship between all species of *Blastomycoides*. None of the species can use nitrates or nitrites, but they all use ammonium salts. It is very characteristic for the *Blastomycoides* that they use spermine, which is a very important fact for the differentiation between *Blastomycoides* and other yeasts or yeast-like filamentous fungi. As to sugars the one most preferred seems to be maltose, but lower alcohols as ethanol e.g. are used only scarcely by *Bl. cerolytica*.

The aim of this study was to obtain a controllable conversion of the filamentous growth of *Blastomycoides* into the yeast-like phase of growth. This was completely realised by using different tryptophane desaminase inhibitors. After two to three reinoculation from this growth phase into commonly used media, another filamentous growth was observed. Sexual forms, represented in the genus *Blastomycoides* in the nude, undifferentiated asci, were observed in all species. All *Blastomycoides* seem to be isogame diplonts. Its haplonts are represented in the yeast phase of growth only, for which reason it was not possible to obtain sexual forms from the yeast phase directly without passage through the filamentous phase where probably a copulation of the haplonts occurs.

DISCUSSION

The presence of asci in *Bl. dermatitidis* was first described by MOORE (1933). DE MONBREUN (1934) discussed the value of MOORE's observation, believing that the spheres observed in the quiescent state not are asci but only chlamydospores containing fatty droplets. In the experimental part of this article a method is described which has helped to establish the truth about this matter. After the bursting of the ascus of a hanging drop colony of *Bl. dermatitidis* we have diluted the suspension of the protruded spores and have inseminated them into Wickerham's medium where mycelial growth has been obtained from every single spore. This means that the problem was a histological mistake and a misinterpretation of the staining properties of fatty droplets and acid fast ascospores.

Bl. brasiliensis shows a very interesting form of asci. They show multiple exoascal protrudings where characteristical conidial bodies are observed later. Morphologically they very much resemble the cysts of *Boudierella coronata* J. CONSTATIN 1897. In *Boudierella coronata* the formation of ascospores is due to a condensation of cytoplasm in the budding forms, in *Bl. brasiliensis* this phase occurs only after meiosis, the budding here being not only plasma condensation but also a reduction of the chromosomes. The spores of *Bl. brasiliensis* cannot again give stellated ascospores if they are inoculated as monosporic colonies into the yeast phase supporting media. Only by passing through the filamentous phase can we find again characteristic sexual forms.

Bl. cerolytica asci were studied in a previous paper (MUFTIC, 1957). It is a haplontic yeast. Copulation occurs only from the filamentous growth phase, a mass of pseudohymenium with asci being formed from ascogenic hooks. It was characteristic for this species that special biochemical factors are needed to accelerate the formation of the dicaryophase. This cytochemical genetical characteristic might be present in other fungi, but has not yet been studied so far.

Bl. immitis seems to have diplobiontic yeast-forms and in this respect differs from the other *Blastomycoides*. CONVERSE (1957) succeeded in obtaining an ascospore-asci-ascospore reproductive cycle by several serial transfers. But it is not necessary that ascospores of *Bl. immitis* multiply by a sexual cyclus. The photographs show that ascospores can be transformed to yeast cells which can grow further into budding forms as shown in the electron-microscope fig. 4. both cells being able to multiply and form asci without the necessity to pass through the filamentous phase. This observation might be considered as still questionable because of a monosporic colony of *Bl. immitis* not yet having been studied.

Considering all morphogenetic characteristics of cytological elements in *Blastomycoides* we may draw the following conclusions: The vegetative forms of the *Blastomycoides* very much resemble each other and show characteristic aspects observed in the Aleurismaceae. Because of this resemblance BOGLIOLO & AROEIRA-NEVES (1952) proposed to classify them as Aleurismaceae. As a matter of fact, all species which are supposed to be Aleurismaceae have yeast-forms which in parasitic circumstances and in special cultural conditions continue to grow as such forms exclusively. This dimorphism which is determinated by certain biochemical factors is the main reason for our opposition against the classification of *Blastomycoides* as Aleurismaceae. Another very important fact is the existence of perfect sexual forms in all species of *Blastomycoides*, forms which represent the simplest of the Ascomyces, i.e. free asci, adjacent to the mycelium or intermingled into a kind of plectenchyme simulating a pseudohymenium. From the mycological point of view and in regard to the sexual forms which have always been decisive characteristics in mycological determinations, I assume that the *Blastomycoides* are very near to the Protomycetaceae and a transitory form between the Endomycetales and the Taphrinales.

In future we will probably be able to classify them as Endomycetales or Taphrinales, but for the moment we can say they are filamentous yeast forms showing fungi with perfect sexual forms which doubtless are ascospores.

SUMMARY

Studies were made on some environmental factors promoting yeast-phase growth of *Blastomycoides* in chemically defined media. With inclusion of different tryptophane desaminase inhibitors, pure yeast phase of growth was obtained. The same factors facilitate the development of perfect sexual forms and the observation of the cytological characteristics. Therefore, *Blastomycoides*, represented by *Blastomyces dermatitidis, Bl. brasiliensis, Bl. cerolytica* and *Bl. immitis* (formerly *Coccidioides immitis*), form a special genus which is very near to the Protomyces and represents a transitory phase between the Endomycetales and the Taphrinales. Although the vegetative mycelium of the Blastomycoides resembles the Aleurismaceae, they cannot be classified as such, especially because they are dimorphic fungi (filamentous-yeast) and their sexual forms are characteristic asci on a very primitive level. *Blastomycoides* not only represent a group of fungi causing grave visceral mycoses (blastomycoses) but they also are an indiscutable mycological entity, as yet represented in the genus *Blastomycoides*

Bibliography

1. Baker, E. E. & Mrak, E. M.: (1941) Spherule formation in culture by Coccidioides immitis. Rixford and Gilchrist, Amer. J. Trop. Med., **21,** 589—594
2. Bogliolo, L. & Aroeira-Neves, J.: (1952) Researches on the etiological agents of the American blastomycosis. Mycopathol et Mycol. appl. **6,** 137—160.
3. Bullen, J. J.: (1949) The yeast like form of Cryptococcus farciminosus (Rivolta): (Histoplasma farciminosum). J. Pathol. Bacteriol. **61,** 117—120.
4. Burke, R. C.: (1951) In vitro cultivation of the parasitic phase of Coccidioides immitis. Proc. Soc. Exp. Biol., N.Y. **76,** 332—335.
5. Conant, N. F.: (1941) A cultural study of the life-cycle of Histoplasma capsulatum Darling 1906, J. Bacteriol. **41,** 563—580.
6. Conant, N. F. & Vogel, R. A.: (1954) The parasitic growth phase of Coccidioides immitis in culture. Mycologia **46,** 157—160.
7. Constantin, J.: (1897) Sur une entomothorée nouvelle. Bull. Soc. mycol. France, **13,** 38—43.
8. Converse, J. L.: (1955) Growth of spherules of Coccidioides immitis in a chemically defined liquid medium. Proc. Soc. Exp. Biol. N.Y., **90,** 709—711.
9. Converse, J. L.: (1957) Effect of Surface Agents on Endosporulation of Coccidioides immitis in a Chemically defined medium. J. Bacteriol. **74,** 106—107.
10. Cook, E. and others: (1939) The effect of 1, 2, 5, 6-dibenzanthracene on the growth and respiration of yeast. Amer. J. Cancer, **35,** 543—545.
11. Demonbreun, W. A.: (1934) The cultivation and cultural characteristics of Darlings Histoplasma capsulatum. Amer. J. Trop. Med. **14,** 93—125.
12. Demonbreun, W. A.: 1935) Experimental chronic cutaneous blastomycosis in monkeys. A study of the etiologic agent. A.M.A. Arch. Derm. & Syphil. **31,** 831—854.
13. Dodge, C. W. & Sanford, B.: (1937) Some effects of metal cholanthrene on

the morphology and growth of yeasts. Missouri Bot. Gard. Ann., **24,** 583—590.
14. Fialho, A.: (1947) A impregnaçao argentica nas investigaçoes micologicas. Arch. histol. B. Air., **3,** 443—463.
15. Lack, A. R.: (1938) Spherule formation and endosporulation of the fungus Coccidioides immitis in vitro. Proc. Soc. Exp. Biol. N.Y. **38,** 907—909.
16. Levan, A. & Sandwall, C. G.: (1943) Quantitative investigations on the reaction of yeast to certain biologically active substances. Hereditas, Lund, **29,** 164—178.
17. Lubarsky, R. & Plunkett, O. A.: (1955) In vitro production of the spherule phase of Coccidioides immitis. J. Bacteriol. **79,** 182—186.
18. Mackinnon, J. E. & Vinelli H.: (1950) Caracteres diferenciales de paracoccidioides brasiliensis y blastomyces dermatitis en los Tejidos; utilidad de la coloracion con el carmin de Best. An. Fac. med. Montevideo, **35,** 299—310.
19. Macneal, W. J. and Taylor, R. M.: (1914) Spherule formation in vitro. J. med. Res. **30,** 261—269.
20. Mickle, W. A. & Jonnes C. P.: (1940) Dissociation of Candida albicans by lithium chloride and immune serum. J. Bact., Balt., **39,** 633—647.
21. Moore, M.: (1932) Coccidioidal granuloma: classification of the causative agent (Coccidioides immitis). Missouri Bot. Gard. Ann. **19,** 397—428.
22. Moore, M. (1933) Blastomycosis: report of a case with a study of an etiologic factor and a classification of the organism. Missouri Bot. Gard. Ann. **20,** 79—118.
23. Muftic, M. (1957) Blastomyces cerolytica (sp. n.) and its relation to Coccidioides. Mycopathol. et Mycol. Appl., **8,** 18—26.
24. Nadson, G. A. & Krassilnikov, N. A. (1932) Stroenie razvitie i rassobrazovanie u endomitzetov (endomycetaceae). C. R. Acad. Sci. URSS, ser. A., **13,** 313—322.
25. Negroni, P.: (1949) Estudios sobre el coccidioides immitis Roxford. An. Soc. Sci. Argent. **148,** 333—342.
26 Negroni, P. & Daglio, G. A.: (1949) Aplicacion de nueva tecnicas para el estudio fisiologico de los hongos levaduriformes. An. Soc. Sci. Argent., **148,** 271—278.
27. Nickerson, W. J. & Van Rij, N. J.: (1949) The effect of sulfhydryl compounds penicillin and cobalt on the cell division mechanism of yeasts. Biochim. biophys. acta, **3,** 461—475.
28. Punkari, J. & Henrici, A. T.: (1933) A study of variation in a chromogenic asporogenous yeast. J. Bact., Balt. **26,** 125—138.
29. Rondoni, P. & Barbieri, G. P.: (1950) The activation of some carcinogenic compounds an SH activated enzymes. Enzymologia, **14,** 10—14.
30. Scherr, G. H.: (1952) Studies of dimorphism mechanism in Saccharomyces cerevisiae. Mycopathol. et Mycol. Appl. **6,** 182—230.
31. Segal, R. B.: (1939) Vliianie vysshikh spirtov na drozhi'zhi. Mikrobiologiia, Moskva, **8,** 466—469.
32. Titsworth, E. H. & Grunberg, E.: (1950) A medium for the growth and maintenance of the yeast-like phase of histoplasma capsulatum. Mycologia, Lanc., **42,** 298—300.
33. Zikes, H.: (1920) cited according reference No 2.
34. Vuillemin, P.: (1919) La genre blastomyces luteus en synnonimie avec Aleurisma flavissimum. Bull. Soc. Sci. Nancy, **3,** 151—160.

INVESTIGATIONS INTO THE ROLE OF FUNGI IN PULMONARY DISEASES

by

H. S. Andleigh, M.D., D.T.M., D. Bact. (Lond.)

Reader in Pathology & Bacteriology, S.M.S. Medical College, Jaipur, India

(30.IX.1959)

The study of Pulmonary Mycoses is of considerable practical interest, since diseases caused by fungi, or in which fungi may be suspected as etiologic agents, constitute a sizeable percentage of the disorders affecting the lungs. The incidence of Pulmonary mycoses seems to be increasing day by day. This is not only due to the improved diagnostic methods but also because the clinicians have started taking more and more interest in medical mycology. Drugs come as a mixed blessing and so do the chemotherapeutic agents and antibiotics because by subduing bacteria they allow the free growth of fungi in the body. The subject was a neglected one in the past and much progress could not be done, but as the physicians and the pathologists are becoming more fungus conscious, more and more cases are coming to light. However, their mode of infection, whether from exogenous or endogenous source and their pathogenesis is still not very clear. Moreover, the study of mycotic infections of the lungs makes up one of the most interesting chapters in the field of nontuberculous diseases of the chest. Cough, mucoid or purulent expectoration, occasional hemoptysis, pleural pain, loss of weight, night sweats and gradual weakness as found in tuberculosis are also the main symptoms of the pneumomycosis like Moniliasis Cryptococcosis, Histoplasmosis, Geotrichosis and Actinomycosis. In their close resemblence to tuberculosis both as to the symptomatology and radiological findings, the Pneumomycosis offer a challenge to the diagnostic ability of the internist, chest specialist and radiologist.

We undertook these investigations to find out the role played by fungi in Pulmonary diseases and whether the fungus isolated was the primary or a secondary invader. Cases suffering from chronic pulmonary diseases like bronchiectasis, lung abscess, tuberculosis and malignancy lung not showing any beneficial effect of the routine treatment given to them were selected for investigations. These cases were divided in two groups.

1. Non-tuberculous chronic pulmonary diseases.
2. Pulmonary tuberculosis with sputum positive for A.F.B.

In the first group the sputum was subjected to the following investigations:

1. (a) Direct Examination.
    - i) 4% KOH preparation.
    - ii) Gram's staining
    - iii) Ziehl Neelsen's Staining for A.F.B.
    - v) a) Without concentration
        - b) With concentration.
    - iv) P.A.S. Stain.

    (b) Cultural investigations on the following media:
    - i) Sabouraud's medium with Achromycin.
    - ii) Sabouraud's medium with Achromycin and Actidione.
    - iii) Brain Heart Agar with Achromycin.
    - iv) Brain Heart Agar with Achromycin and Actidione.
    - v) Thioglycollate medium.

Achromycin was added to prevent the growth of bacterial flora and Actidione to check the growth of non-pathogenic fungi.

In the second group, the sputa in addition to the above investigations were also cultured on the following media:

- i) Lowenstein Jensen medium plain.
- ii) Lowenstein Jensen medium with 20 microgram of Streptomycin per ml. of the medium.
- iii) Lowenstein Jensen medium with 100 micrograms of streptomycin per ml. of the medium.

Streptomycin was added to the medium to find out if the tubercle bacilli had become resistent to streptomycin and thus keeping up the infection.

Sensitivity of the organisms isolated was tested against Nystatin. Brain heart liquid medium was used as the culture medium for carrying out these sensitivity tests. Serial dilution method as used by me for testing the aromatic diamidines (ANDLEIGH, 1957) was used to carry out the sensitivity tests. All the strains of *C. albicans* isolated were sensitive to Nystatin. No growth of any strain of *C. albicans* isolated was obtained in a concentration of 50 microgram per ml of the medium. The strains of *Actinomyces bovis* were not found to be sensitive to Nystatin. The following observations were made in the first thirty two cases.

(GROUP I) TABLE I.

		Total No. of cases	Positive for fungi or A.F.B. on microscopic Examination
1.	4% KOH	32	4 positive for fungi.
2.	Gram's Staining	32	6 positive for fungi.
3.	P.A.S. Staining	32	6 positive for fungi.
4.	Ziehl Neelsen's staining for A.F.B.	32	
	i) Without concentration		None positive for A.F.B.
	ii) With concentration.		None positive for A.F.B.

(GROUP I) TABLE II.

S. No.	Disease (Provisionally diagnosed)	No. of cases	No. of positive cases for fungi	Type of fungi grown
1.	Lung abscess	17	4	C. albicans.
2.	Bronchiectasis	5	2	C. albicans.
3.	Chronic bronchitis	8	2	C. albicans.
4.	Malignancy Lung	2	1	Actinomyces bovis.

All these cases were positive for fungi even on repeated cultural examination. Fungi were isolated from their bronchial aspirates also. Two of the patients from whom *C. albicans* was isolated from sputum on repeated cultures and from their bronchial aspirates also were treated with Mycostatin tablets. Two tablets were given three times a day for seven days. The patient improved rapidly on Mycostatin.

GROUP II (TUBERCULOUS) TABLE III.

No. of cases	Positive for A.F.B. Smear Exam.	Positive for fungi (Culture)	Plain Lowenstein Jensen Medium	20 μgm Streptomycin per cc of Lowenstein Jensen Medium	100 μgm Streptomycin per cc of Lowenstein Jensen Medium
10	10	2 cases *C. albicans*)	10 cases positive for A.F.B.	No growth	No growth

The following conclusions were drawn:

1. Out of 32 cases of non-tuberculous chronic pulmonary diseases 9 cases were positive for fungi. Out of them *C. albicans* was isolated from eight cases and from one case *Actinomyces bovis* was isolated. They were isolated from the sputum on repeated cultural examination and from the bronchial aspirates also.
2. Out of the 10 cases of pulmonary tuberculosis two were positive for *C. albicans.* The acid fast bacilli isolated from these cases were sensitive to Streptomycin. It can, therefore, be concluded that in some cases of pulmonary tuberculosis, suspected of infection with a resistent strain, the disease is kept up due to a superadded fungal infection and not due to any resistance developed by the organisms against streptomycin.
3. All the strains of *C. albicans* isolated were sensitive to Nystatin in a concentration of 50 microgram per ml of the medium and two cases treated with Mycostatin tablets improved rapidly.

SUMMARY

32 cases of non tuberculous chronic pulmonary diseases and 10 cases of chronic pulmonary tuberculosis not responding to the routine treatment, were investigated to find out if fungi played any role in their etiology. Fungi were isolated from 9 cases of non-tuberculous and 2 cases of tuberculous pulmonary infections.

Fungi as such seem to play some role in the etiology of Pulmonary Diseases and probably also help in keeping up the disease in tuberculous cases. Mycostatin is the drug of choice in the treatment of pulmonary moniliasis.

Sputa and bronchial aspirates from 32 cases of non-tuberculous chronic pulmonary diseases and 10 cases of chronic pulmonary tuberculosis, not responding to the routine treatment, were repeatedly examined to find out if fungi played any role in their etiology. Fungi were isolated from 9 cases of non-tuberculous (*C. albicans* from 8 and *Actinomyces bovis* from 1) and two cases (*C. albicans* from both) of tuberculous pulmonary infections. All the strains of *C. albicans*and *A. bovis* isolated were tested for their sensitivity against Nystatin. Only strains of *C. albicans* were found to be sensitive to Nystatin. Two cases treated with Mycostatin improved rapidly.

ZUSAMMENFASSUNG

Sputum and Bronchialsekret von 32 Fällen von nicht-tuberkulösen chronischen Lungenkrankheiten und von 10 Fällen von chronischer Lungentuberkulose, die auf Routinebehandlung nicht ansprachen, sind wiederholt daraufhin untersucht worden, ob Pilze eine aetiologische Rolle spielten. Pilze sind in 9 nicht-tuberkulösen Fällen (*C. albicans* in 8, *Actinomyces bovis* in einem Falle) und in 2 tuberkulösen Fällen (*C. albicans* in beiden) isoliert worden.

Alle isolierten Stämme von *C. albicans* and *A. bovis* sind auf

ihre Sensitivität gegen Nystatin untersucht worden. Nur *C. albicans* war sensitiv gegen Nystatin. Zwei Fälle, die mit Mycostatin behandelt wurden, zeigten rasche Besserung.

Acknowledgements

My thanks are due to Messers Sarabhai Chemicals for kindly supplying the Nystatin and Mycostatin tablets.

References

1. Andleigh, H. S., (1951) Laboratory Diagnosis of Mycotic Lung Infections. J. Ind. med. Ass. **XX**, 10, 360—371.
2. Andleigh, H. S. (1957) In vitro study of antifungal activity of Pentamidine and Stilbamidine. Mycopathol. Mycol. Appl. **7**, 135—137.
3. Andleigh, H. S. (1957) Fungicidal activity of Aromatic diamidines. J. Ind. med. Ass.
4. Andleigh, H. S. (1958) Investigations into the role of fungi in Pulmonary Diseases in India. Amer. Rev. Tuberculosis **78**, 644—646.
5. Heilig, R. E. & Andleigh, H. S. (1958) Broncho-Pulmonary Moniliasis. J. Ind. med. Ass. 30, **3**, 82—90.
6. Kasliwal, R. M. & Andleigh, H. S. (1954) An interesting case of Broncho-Pulmonary Moniliasis. Ind. J. med. Sci.
7. Kligman, A. M. (1950) Aids in Technic in the Identification of Candida albicans. J. Invest. Dermat. **14**, 173.
8. Lipnik, M. J., Kligman, A. M. & Strauss, R. (1952) Antibiotics and Fungous Infections. J. Invest. Dermat. **18**, 247.
9. Oblath, R. W., Donath D. H., Johnstone, H. B. & Kerr, W. J. (1951) Pulmonary Moniliasis. Arch. Int. Med. **35**, 97.
10. Weed, L. A., Baggenstoss, A. H. & Bangher, L. (1949) Some problems in the diagnosis of Actinomycosis. Proc. Staff. Meet. Mayo Clinic **24**, 463.

MYCOLOGICAL NOTES.
NEW SPECIES, COMBINATIONS, AND HOST RECORDS

by

MARIE L. FARR[1]

(18.XII.1959)

Cercospora antigononis sp. nov.

Several species of *Cercospora* have been described from various members of the family Polygonaceae — chiefly on *Rumex* and *Polygonum* — but, as far as can be ascertained from available records, none have been reported infecting *Antigonon*. Some dried *A. leptopus* HOOK & ARN. leaves recently received showed infection by a *Cercospora*. The disease was manifested mostly by individual light-brown, bordered spots, but on some leaves large areas surrounding the spots had turned brown, and on those areas the fruiting portion of the fungus was most copiously developed. Perhaps, however, a species of *Periconia*, also abundant in those areas, was partly responsible for the extensive discoloration. According to the label accompanying the specimen, the disease occurs during the winter months only, and results in leaf drop soon after infection.

CHUPP'S monograph (CHUPP, Charles. 1953. A monograph of the Fungus Genus *Cercospora*. 667 pp., illust. Ithaca, New York.) accepts a dozen or so definite species known to infect members of the Polygonaceae, but the fungus here described fits none of these and Prof. CHUPP agrees that it is an undescribed species. He states (personal communication) "the pathogen is a *Cercospora* with cylindric, pale olivaceous conidia that are more nearly permanently catenate than any other species I studied."

Cercospora antigononis sp. nov. (fig. 1)

Maculae distinctae, fere orbiculares vel angulatae vel irregulares, 1—9 mm. diam., pallide brunneae margine fusco vel nigro; caespituli amphigeni, praecipue hypophyll; stromatica carentia vel minuta; conidiophora fasciculata, divaricata vel compacta, densa, pallidebrunnea, recta vel leviter undulata, rare geniculata, simplicia,

---

[1] Crops Research Division. Agricultural Research Service. United States Department of Agriculture. Beltsville, Maryland.

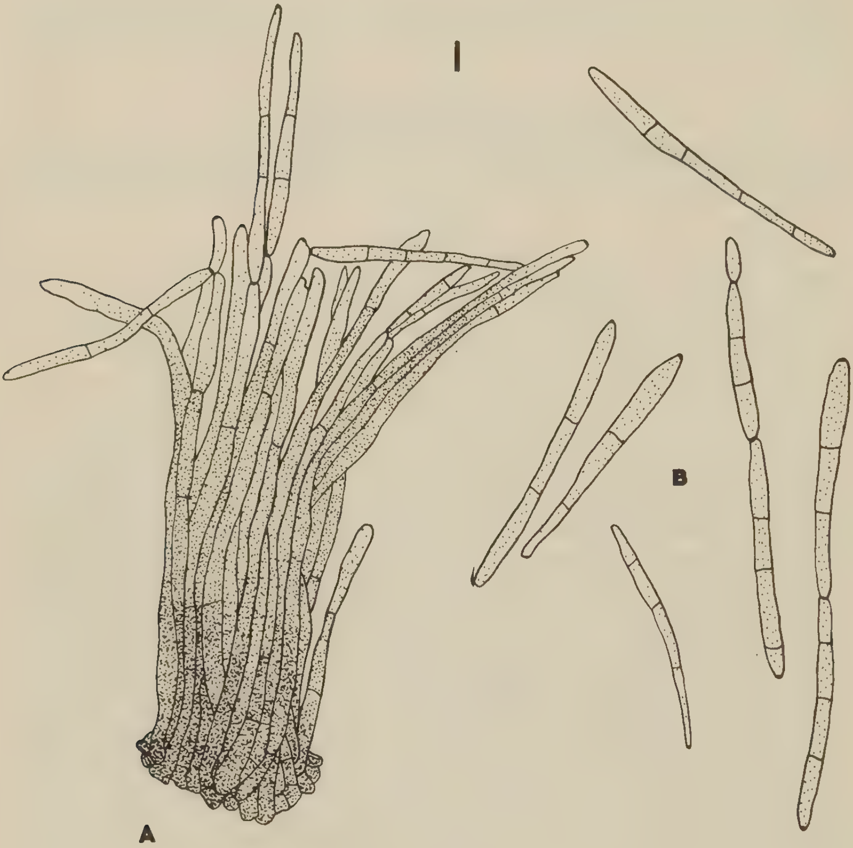


Fig. 1. *Cercospora antigononis* sp. nov.: A, habit; B, conidia, some in chains; all c. × 476.

septata, 70—190 × 3—4μ, ad apicem pallidioria, leviter attenuata vel amplificata, obtusa vel rotundata vel subtruncata cicatricio praedita; conidia subhyalina vel pallide olivacea, plerumque catenulata, cylindracea, 0—ad 3—(4—) septata, recta, cicatricio conspicuo ad locos affictionis praedita, ad basim obtusa vel subacuta, ad apicem obtuse rotundata vel subtruncata, 15—84 × 4.5—6μ. In foliis vivis *Antigononis leptopi* HOOK. & ARN., Texas, U.S.A.

Spots amphigenous, light-brown, usually with dark-brown to blackish margin, round, angular, or irregular, 1—9 mm. diam.; fruiting amphigenous but predominantly hypophyllous; stromata lacking or consisting of a small group of brown cells; conidiophores in dense (sensu CHUPP), divergent to coremioid fascicles, light-brown with paler tips, straight to slightly undulate in the upper part, simple, septate, rarely geniculate, even but sometimes with

undulating margins and attenuate or slightly enlarged tips, which are obtuse, rounded, or subtruncate with a spore scar, 70—190 × 3—4μ; conidia subhyaline to pale olivaceous, mostly in chains of 2—3, mostly narrowly cylindric, but varying to narrowly obclavate, straight, 0— to 3(4—) septate, 15—84 × 4.5—6μ, with a conspicuous attachment scar at one or both ends, the apex bluntly rounded to subtruncate, the base blunt, obtuse to subacute.

TYPE: On living *Antigonon leptopus* Hook. & Arn. leaves, Brownsville, Texas, U.S.A., 15. March 1959, Runyon Herbarium, Cryptogamae no 4641 (type), coll. Robert Runyon.

Cercospora dioscoreae Ell. & Mart. in Guatemala (fig. 2).

A rapidly spreading *Cercospora* leaf spot disease in Guatemala merits special notice, as it is causing serious concern in commercial yam plantations. The hosts involved are *Dioscorea composita* Hemsl. and *D. floribunda* Mart. & Gal.

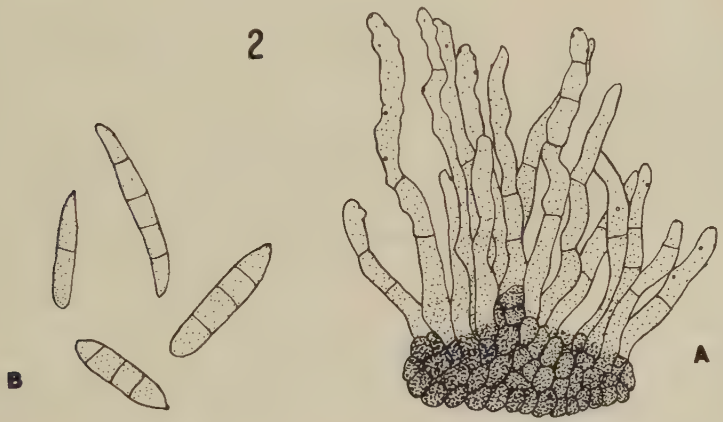


Fig. 2. *C. dioscoreae* on *Dioscorea composita* Hemsl.: A, habit; B, conidia, all c. × 650.

Because of its rather unusual appearance, the fungus was at first thought possibly to be a new species, but has been identified by Chupp as *C. dioscoreae*. Leaf collections sent here by Dr. Ernest P. Imle of the American Cocoa Research Institute show a striking pattern of yellow blotches on which the dark brown stromata of the fungus form extensive colonies and are readily visible with the unaided eye.

This appears to be the first report of *Cercospora dioscoreae* from Guatemala and on these species of *Dioscorea*.

Colletotrichum oca sp. nov.

Relatively few fungi have been reported from hosts belonging to the family Oxalidaceae, and no species of *Colletotrichum* has been found among these. The host plant from which the present new species is described is locally known as ,,oca'' and is of some importance in Bolivian country districts, where the boiled tubers constitute part of the people's diet.

This fungus appears to be closest morphologically and in measurements to *Colletotrichum fusarioides* (ELL. & KELL.) O'GARA (on *Asclepias*) and to *C. phyllachoroides* (ELL. & EV.) v. ARX (on *Artemisia*), but as its host belongs to a different family, it is at present considered a distinct species.

Colletotrichum oca sp. nov.

Maculae epiphyllae, e viride pallentes vel laete brunneae margine pallide viridi, orbiculares, ad 2 mm. diam.; acervuli solitarii vel gregarii, fusci vel nigri, 150—450μ diam., setosi, plusminusve pulvinati, irregulariter orbiculares ad elongati; setae laete vel atrobrunneae, continuae, leviter curvatae, 60—100 × 4.5—8μ; conidiophora irregulariter cylindracea, dilute brunnea ad apicem pallidiora, ad 25μ longa; conidia subhyalina vel fumosa, continua, subrecta vel leviter curvata, cylindracea vel subcylindracea, utrinque rotundata vel obtusa, (17) 21—25 (26) × 4.5—6μ. In foliis vivis *Oxalidis tuberosae*, Bolivia.

Spots epiphyllous, pale green or light brown with a pale green border, circular, up to 2 mm. diam.; acervuli solitary or gregarious, dark brown or black, c. 150—450μ diam., setose, more or less pulvinate, irregularly rounded to elongate in outline; setae light brown to blackish, continuous, slightly curved, 60—100 × 4.5—8μ; conidiophores irregularly cylindrical, light brown with paler tips, to 25μ long; conidia subhyaline to smoky, continuous, straight to slightly curved, cylindrical to subcylindrical, the ends rounded or broadly obtuse, (17) 21—25 (26) × 4.5—6μ.

TYPE: On living *Oxalis tuberosa* leaves, La Paz, Bolivia, April 1955, Cochabamba Est. Exp. Agr. no. 186, coll. Segundo Alandia B.

Aposphaeria oxalidis sp. nov.

Pycnidia superficialia, caespitosa vel adnata, sessilia, contextu carbonaceo, pseudoparenchymata, glabra, globosa vel subglobosa, vertice in ostiolum conoideum brevissimum attenuato, 240—260μ diam.; ostiolo crasso, nigro, periphysato, aperturo 20μ diam.; conidia continua, hyalina, oblonga, 3—4 × 1.5μ. In tuberis *Oxalidis tuberosae*, Bolivia.

Pycnidia superficial, closely gregarious to crowded, sessile, carbonaceous, the wall made up of polygonal pseudoparenchymatous cells 20—30μ diam., glabrous, globose or subglobose, ostiolate, 240—260μ diam.; ostiole slightly conoid, with a thickened, black, periphysate rim, the opening 20μ diam.; conidia continuous, hyaline, oblong, 3—4 × 1.5μ.

TYPE: On tuber of *Oxalis tuberosa*; Cochabamba, Bolivia, June 1956, Cochabamba Est. Exp. Agr. no. 211, coll. Segundo Alandia B.

Septoria malvicola Ell. & G. Mart. on a new host in a new locality.

Septoria malvicola Ell. & G. Mart. has been reported, as far as can be determined, on two genera of Malvaceae, *Althea* spp. and *Malva* (*M. rotundifolia*). Its known geographic distribution is predominantly in the northeastern and middle west states, but extending west to Texas and Oklahoma.

Recently I examined a large collection of dried *Sphaeralcea cuspidata* (A. Gray) Britton leaves collected at Bernalillo, New Mexico, in 1907, and infected by a *Septoria*. Comparison with named *Septoria malvicola* specimens showed the fungus under consideration to be identical with that species. It also corresponds well with the description, being characterized by a largely epiphyllous habit; blackish, depressed, membranous perithecia 80—110μ diam.; and conidia 33—37μ long. This previously overlooked herbarium material thus represents a new host, as well as a western extension of geographical range, for *Septoria malvicola*.

Septoria cerastii Desm. & Rob. on *Dianthus*

S. cerastii Desm. & Rob., occurring on *Cerastium* (Caryophyllaceae) is easily distinguished morphologically from the various *Septoria* spp. reported from *Dianthus* (Caryophyllaceae) by its distinctly narrower (1μ wide) and often longer conidia. A *Septoria* with conidia 25—45 × 1—1.5μ and indistinguishable morphologically from *S. cerastii* has been found causing a leafspot of a plant designated as „garden pink (*Dianthus plumarioides*)", collected in Jacksonville, Texas, by P. A. Young, May 29, 1959.

Phomopsis phaseoli on *Hordeum*

Apparently the only *Phomopsis* spp. reported so far from any grass host are *P. achilleae* (Sacc.) Hoehn. on barley (Lebedova, L. A. 1933. Fungi et Myxomycetes Kareliae Rossicae. Trudy Bot. Inst. Akad. Nauk. SSSR, ser. 2, no. 1 : 385.) and *P. phaseoli* (Desm.) Grove on *Zea* (Wehmeyer, L. E. 1933. The genus *Diaporthe* Nitschke and Its Segregates).

An agar culture recently received at the National Fungus Collections and originating from barley seeds in Virginia developed into a *Phomopsis* with abundant α & β spores exuding in cream-colored gelatinous masses from the pycnidia. Spore measurements (ave. α : 4—6 × 2—2.5μ; ave. β: 17—22 × 1μ) and other characters of this fungus generally fit the description of *P. phaseoli* as presented by Wehmeyer and by Harter (J. Agric. Res. 11(10): 481—843. 1917). The spore dimensions of *P. achilleae* are too large to accommodate this fungus.

Dimeriella erysipheoides (Ell. & Ev.) comb. nov.

Basonym: *Dimerosporium erysipheoides* Ell. & Ev., J. Mycol. 4(12) : 121. 1888. (Type: Ell. & Ev. No. Amer. Fungi 2341).

A study of *Dimerosporium erysipheoides* type material shows this fungus to be a *Dimeriella.*

The genus *Dimerosporium* FCKL. was originally based on a fungus later found to be an *Asterina* and is therefore a synonym of the latter; but because of a misstatement in SACCARDO'S (I : 51) description of the fruiting bodies FUCKEL'S genus eventually became a mixture of asterinaceous, sphaeriaceous, and perisporiaceous fungi (see discussion by v. HOEHNEL, 1910, Frag. Mykol. n. 477).

Recently collected material on *Cyonodon* from Cambodia (Litzenberger 82) is identical with type of the ELLIS & EVERHART fungus, but is in better condition and contains numerous asci, ascospores, and hyaline, filiform paraphyses.

Dimeriella erysipheoides (ELL. & EV.) comb. nov. is distinct from *Dimeriella disseminata* SYD. and *Dimerosporium dubiosum* SPEG. both on Gramineae).

The drawings were made with the aid of a Bausch & Lomb camera lucida. Types of the new species are deposited in the National Fungus Collections, Beltsville, Maryland, U.S.A. Duplicate type material of *Cercospora antigononis* is in the herbarium of the Department of Plant Pathology, Cornell University, Ithaca, New York.

I should like to express my appreciation to Mr. JOHN A. STEVENSON for his interest in this study; Prof. CHARLES CHUPP for examining the *Cercospora* species; Miss EDITH K. CASH and Dr. SYDNEY F. BLAKE for correcting the Latin diagnoses; and Dr. ERNEST P. IMLE and Mr. PAUL RUSSELL for supplying information about *Cercospora dioscoreae* ELL. & MART. and *Oxalis tuberosa* MOL. respectively.

MEDICAL MYCOLOGY IN SCANDINAVIA AND FINLAND DURING A CENTURY, FROM 1841 TO 1940.

by

HEITI PALDROK, M.D.

From the Department of Dermatology, Karolinska Sjukhuset, Stockholm 60, *Sweden. Head: Professor* SVEN HELLERSTRÖM, *M.D.*

(3.XI.1959)

INTRODUCTION

The traditions of Medical Mycology in Scandinavia and Finland are closely connected with the very beginning of this branch of medical science on the European continent.

Two Swedish physicians, Frederik Theodor BERG and Per Henrik MALMSTEN, happened to visit the Paris hospitals in 1840/41 (BERG (S)*), 1842 a, 1846 a, b) and 1842/43 (LENNMALM (S), 1911) respectively, that is at the time of the first discoveries that plant parasites could cause diseases in human beings. Enthusiastic about the aspects the new discoveries offered, BERG and MALMSTEN, after having returned to Stockholm, continued to work on these problems.

BERG (S), being a pediatrician, devoted himself to the study of the occurrence of a yeast-like organism in thrush in children, and as a result of these studies, he, in 1841, declared thrush to be an independent clinical entity caused by a yeast-like fungus.

This statement by BERG was, in 1842, also quoted by Johannes MÜLLER (Berlin), who had been present when BERG first introduced the subject before the Swedish Medical Society in Stockholm in September, 1841 (MÜLLER, 1842, BERG 1846 a, b).

Carrying on his studies (1842 d, 1843), BERG then, in 1846, published more complete data on the subject, including, inter alia, reports on the cultivation results, as well as fermentation studies with the causative agent, but also experimental transfer of the disease to healthy infants.

As far as we can judge from literary remains at hand, BERG (S) made his statements independently of LANGENBECK'S discovery, in 1839, of a yeast-like fungus in the post-mortem of a patient, dead of

---

*) The letters D, F, N, S in parenthesis after the authors' names indicate their country of origin: Denmark, Finland, Norway, and Sweden.

Typhus abdominalis, which fungus LANGENBECK (Germany) assumed to be the cause of this ailment. The priority of BERG (S) in the recognition of a yeast-like fungus, known today as *Candida albicans*, as the cause of thrush has in later years many a time been called to attention by Scandinavian scientists (FLENSBURG (S), 1916).

About the same time as BERG (S) worked in Stockholm, in Copenhagen, Denmark, Adolph HANNOVER (D) — who, in 1839 had shown an alga to be capable of causing pathological changes in a triton, viz., *Triton punctatus* — having just returned from Berlin, where he had been working with Johannes MÜLLER (FIBIGER (D), 1915), undertook a control of LANGENBECK'S statements. HANNOVER (D) encountered the yeast-like fungus in a series of postmortems, but also on living human beings with different kinds of disorders, and came to the conclusion that the fungus was only a secondary invader in the organisms of the patients, where the power of resistance was low on account of the primary disease.

This work was published in 1842. In 1847 HANNOVER (D) critically reviewed BERG'S (S) monograph on thrush in children issued in 1846, and added to the review his own views concerning the nature of the affection. Whereas BERG (S) had interpreted thrush to become at times a rather serious disease, HANNOVER (D) looked upon it as a disturbance of local character only, not causing any tissue damage, nor death of the afflicted individual.

From Finland ILMONI (F), in 1842, in a communication on Diabetes mellitus, delivered at the Meeting of Scandinavian Naturalists, mentioned the occurrence of yeast-like fungi in diabetic urine.

In February, 1844, MALMSTEN (S) observed in Stockholm a case of hair infection caused by a fungus that differed from GRUBY'S *Microsporum*, or Phyto-alopecia. That fungus, besides invading the hair, also attacked the roots of the hair. MALMSTEN (S) had made this observation independently of GRUBY (Paris) but he had early in the spring of the same year discussed the matter with the latter by correspondence (MALMSTEN (S) 1845 b, c, 1848).

GRUBY was the first to announce the new observation (April 1st, 1844), whereas MALMSTEN (S) came briefly after with a communication (1844 a), and then a closer description of the condition, followed by an illustration of the invaded hair (1844 b).

Both the authors called the new clinical picture for Rhizophyto-alopecia. MALMSTEN (S), however, in 1845 (b, c), changed its name to Trichophyton tonsurans, a designation which is still in general use.

GENERALITIES

The striking discoveries in the field of clinical mycology mentioned in the introduction, stirred the minds of many a bacteriologically or botanically minded medical man in Scandinavia. However, they gave, on the other hand, also rise to speculations of different kinds. In 1851 e.g. ARNDTSEN (N) claimed a *Puccinia* species, whose conidia he had observed microscopically in crusts from lesions in the capillitium of a patient with Favus, to be the cause of the trouble. Those statements by ARNDTSEN (N) were in 1852 quoted by W. BOECK (N). But, as seen later from a contribution by BIDENKAP (N) to a debate (1885), the *Puccinia* had got into the lesions only accidentally, from the straw in the patient's bed.

EKLUND (S), in 1883, thought that he was able to confirm LANG'S (Vienna) assumption that Psoriasis was caused by a fungus, *Lepocolla repens* (*Epidermophyton*). EKLUND (S) had microscopically found fungous filaments in scrapings from psoriatic lesions, and stated that he had obtained growth of the fungus after 2-3 days' cultivation in a liquid medium. The author, however, did not determine the fungus closer.

One encouters also studies into the fungous flora of a certain region, or some secretion of the human body. Thus HJELT (F), in 1864, observed that fungi may cause inflammation of the vagina, and RASMUSSEN (D), in 1883, in his thesis on the cultivation of microorganisms from the sputum of healthy man, enumerated a series of moulds, as well as yeasts and yeast-like fungi. He did not, however, analyse the clinical significance of these finds.

There exist also reports on demonstrations of pathogenic fungi before Medical Societies, or at Medical Meetings. C. BOECK (N), in 1885, 1887, and 1894 demonstrated pathogenic fungi and certain methods for their identification in skin lesions, and in 1899 JOHAN-SOPP (N) demonstrated cultures of Herpes tonsurans, both before the Medical Society in Kristiania (Oslo). In 1904 HARBITZ (N) and GEIRSVOLD (N), before the same Society, commented on some exotic Blastomycoses, and demonstrated cultures of the aetiological agents, viz. *Blastomyces dermatitidis*, isolated by RICKETTS, and received by the courtesy of HEKTOEN (Chicago), as well as several strains of SANFELICE'S *Blastomyces neoformans* (*Crytococcus neoformans*), isolated by CURTIS, BUSSE & LEOPOLD. In 1910 H. BANG (D) demonstrated at the meeting of the Northern Dermatological Society cultures of Dermatophytes, encountered in Denmark.

Treatises on the role and the systematical position of the pathogenic fungi, seen from the general bacteriological point of view, have been published by v. DÜBEN (S),1855, HEDENIUS (S),1873, FINNE (N), 1886, and HEIBERG (N), 1887. Also ORLA JENSEN (D), in 1909, in his treatise on the principles of the natural system of bacteria, touches on the position of fungi.

A thesis on the same subject, but considered from a botanical standpoint, was published by JOHAN-OLSEN (N) in 1893. From a further paper by the same author, in 1897, one can comprehend that JOHAN-OLSEN (N), having been a disciple of BREFELD (ALMQUIST (S), 1893), was a suppotrer of the views of the latter on the pleomorphic nature of fungi and related organisms, such as bacteria, and their intermediary forms (Tuberculomyces, Actinomyces etc.).

At the turn of the century, like elsewhere on the continent, the possibilities of fungi playing a role in the aetiology of tumours were also considered in Scandinavia. In 1900 Anna Stecksén (S), and in 1902 V. P. H. Jensen (D) carried out some experimental studies into the aetiology and the histogenesis of neoplasmata respectively, using strains of *Cryptococcus neoformans* for their experiments. Stecksén's (S) investigations were not fruitful with regard to the purpose of the study. Jensen (D), however, who followed the fate of the yeast cells immediately after the inoculation, was able to state that, whilst the injected cells produced a violent acute inflammation with an active emigration of leucocytes, the succeeding generations of the same fungus cells seemed to be acting chiefly in a mechanical way.

Sjöbring (S) declared in 1901 that according to his experiences, certain cell inclusions in tumours, the so-called Plimmer's bodies, had nothing in common with Blastomycetes. Vedeler (N), in 1907, however, thought that he had detected Blastomycetes in blood, and hinted at a connection of his findings with the aetiology of neoplasmata. But no closer explanation followed.

Nevertheless, there exist in fact also descriptions of cases that might have been Mycoses. Thus Quensel (S), in 1929, described the histopathology of a case displaying intestinal ulcerations, in which he had microscopically been able to show double-contoured round to elongate bodies, occasionally of very irregular shape, and situated either within giant cells, or floating freely within the cell infiltration. Quensel (S) mentioned that the bodies reminded of those found by Christiansen (D) in 1922 in intestinal lesions in a case of Mucormycosis in a pig.

In the beginning of the era of medical mycology the clinicians in Scandinavia as well as in Finland, were, in order to gather knowledge on mycotic infections, constrained to resort, besides the scientific publications on the subject, to clinical lectures (Larsen (D), 1844, W. Boeck (N), 1852, Engelsted (D), 1864) or surveys concerning the role of plant parasites in human beings (Hempel (D), 1849, Hempel & Engelsted (D), 1885, 1859, Fries (S), 1867, Trier (D), 1867, Danielssen (N), 1892), but also case descriptions, followed by data on therapy, as given in annual clinical reports and published in current medical journals (Bergh (D), 1869—1885, de Fontenay (D), 1875—1881, E. Pontoppidan (D), 1885 a, Rasch (D), 1898) as well as short presentations of skin disorders (Engelsted (D), 1879, E. Pontoppidan (D), 1885 b, 1890, 1891, 1896). However, as these papers and reports principally treated superficial disorders, such as Dermatophytosis, Pityriasis versicolor, and Erythrasma, for a closer guidance in the topic they still had to take recourse to foreign manuals.

Although at the turn of the century in bacteriological guides for physicians and medical students (A. Holst (N), 1890, 1901, C. Sundberg (S), 1897) some information was given on pathogenic fungi also causing deeper infections, during the period dealt with no mention had been made of mycotic infections in the Northern Textbook of Medicine, a textbook common for the students of all the Scandinavian countries and Finland. Nevertheless, it was in 1905, when RASCH (D) issued his Diseases of the Skin and their Treatment, that in Scandinavia, in a clinical textbook, space was allotted for a systematical review on diseases of mycotic origin.

RASCH (D) enumerated among fungous affections Trichophytic skin diseases (including Microsporia and Trichophytia), Favus, Pityriasis versicolor, Erythrasma, Botryomycosis (a disease of doubtful aetiology), Blastomycoses (according to GILCHRIST as well as BUSSE and BUSCHKE), Actinomycosis, Mycetoma, Carate, Tokela-u Ringworm, and Trichomycosis nodularis. In the second edition of the book, issued in 1909, he added another affection, Sporotrichosis, and to the group of Blastomycoses he assigned a superficial type of disorder, called Oidiomycosis. In the third edition, issued in 1925, the author transferred Tokela-u Ringworm to Trichophytic infections, adding further to this group Tinea albigena and cutaneous affections caused by *Aleurisma*. At the same time he separated from Trichophytia the affection known as Eczema marginatum and caused by *Epidermophyton inguinale*, and called it Epidermophytia. He discarded Botryomycosis from mycotic infections, but took instead up some new diseases, Dermatitis coccidioides, and Scopulariosis. Further, he made mention of rare Mycoses, such as Discomycosis, Hemisporosis etc., among others also Oosporosis, or Nocardiosis.

The next textbook on Skin Diseases and their Therapy, issued by LOMHOLT (D) in 1939, contained a chapter on Dermatomycoses. LOMHOLT (D), who treated the topic from a clinical, symptomatical point of view, grouped the diseases as Trichophytia, Epidermophytia, Microsporia, superficial yeast Mycoses, Erythrasma, Pityriasis versicolor, Favus, Actinomycosis, Sporotrichosis, Blasto-Mycosis profunda, and Granuloma coccidioidale, omitting the rare Mycoses. He broke up the entity of Trichophytosis, separating from it Epidermophytia, the causative agents of which, the *Epidermophyton inguinale* and *Epid. interdigitale*, he declared not to be able to attack hair. He placed Favus, on account of its being rare in Denmark, behind Pityriasis versicolor. Mycotic nail infections, however, were treated separately together with other nail disorders.

But, as the fungous diseases in the textbooks mentioned above had been treated from the dermatological point of view, a closer description of systemic infections and their therapy, actually, only of Actinomycosis, have been given in the Northern Textbook of Surgery. In this book the chapters on that disease were written in 1920 by ROVSING (D), and in 1932 and 1940 by SÖDERLUND (S).

PHYSIOLOGY

Surveys on the fermenting and decomposing activities of microorganisms, but especially of fungi, followed by speculations on the disease producing capacity of these organisms were published in 1868 by FENGER (D), and, 1876, by PANUM (D).

In 1900 WELANDER (S), in order to evaluate heat therapy of Herpes tonsurans capillitii, investigated the heat tolerance in vitro of a *Trichophyton megalosporon*, isolated from hair. The fungus grew well at 29° C, whereas temperatures below 14° C and above 40° C inhibited its growth, but did not necessarily kill it. The resistance of the cultures to unfavourable temperatures depended on the degree of their development when exposed, the more developed ones being more resistant.

JOHAN-OLSEN SOPP (N) declared in 1915 that fungi (moulds and yeasts) as well as bacteria, like all other living beings, are subject to disease. The disease may, above all, be caused by parasites, among which he could also list Actinomycetes, which called forth disease in animals and human beings. The pathological process, being due to antibiosis, may result in colour alterations of the colonies, abnormal forms, and changes in the physiological properties of the micro-organisms, sterility, etc.

LUND (D), in 1921, observed that follicular hormones, viz. Progynon, exerted no effect on the growth of fungi, *Aspergillus niger* and *Saccharomyces cerevisiae.*

GUNDERSEN (N), in 1925 (1925b, 1926a), studying the attitude of fungi (*Achorion Schönleini, Epidermophyton inguinale, Trichophyton violaceum, Tr. faviforme, Penicillium glaucum*) towards GRAM-coloration, found that the fungi are GRAM negative during their evolution phase in the course of optimal vegetative growth, but become GRAM positive during their involution phase, marked by growth inhibition and sporulation. He also pointed out a growth factor requirement for *Tr. faviforme,* the growth substance being encountered in yeast extract and blood.

In 1934 the author showed that the morphology as well as the coloration of *Microsporon fulvum* in culture was highly dependent on the pH of the medium. The vegetative growth optimum lay about pH 7. Sporulation was richest on the alkaline side, and the coloration of the culture varied, according to different H-ion concentrations from yellow to rosy and yellowish brown, until turning white.

In 1940 HALLBERG (S), as well as KRISTENSON (S) staining yeast-like fungi of the Oidium type by HALLBERG'S, and by ZIEHL-NEELSEN'S methods, found these fungi to be partially acid-fast.

DERMATOPHYTOSIS

In Sweden, after MALMSTEN (S) had described the Trichophyton tonsurans in 1844, BERG (S), that same year, and MALMSTEN (S), in 1845 (a), each reported on a case of Favus.

In 1863 TROILIUS (S), on account of a questionnaire from the Administration of Public Assistance in Paris, forwarded to the Royal Medical Board, Stockholm, a report on the occurrence of Favus and Herpes tonsurans in Sweden. It appeared that during the period from 1853 to 1860 in the whole country, and up to 1862 in Stockholm, altogether 350 cases of Favus and 3 cases of Herpes tonsurans had been treated. No expert had been appointed to combat the infection. However, indigents could apply to poor-law doctors for treatment, and, if necessary, due attendance could be afforded to the patients at a hospital in Stockholm.

According to WELANDER (S) (1900) during the period from 1888 to 1899 at the St. Göran Hospital in Stockholm 38 cases of Herpes tonsurans had been treated.

More on the occurrence of Dermatophytosis in Sweden was published in case reports (FRIES (S), 1867; WALDENSTRÖM (S), 1869, 1872; NYSTRÖM (S), 1890; SEDERHOLM (S), 1893; MÖLLER (S), 1902; ALMQUIST (S), 1904; SJÖGREN (S), 1904; STRANDBERG (S), 1913, 1914, 1920; ALMKVIST (S), 1924/25; SCHLASBERG (S). 1929).

STRANDBERG (S), in 1914 in Vienna having followed SCHRAMEK'S investigationsinto dysidrosiform Eczema, in 1917 published a review on a fungous disease on hands and feet.

It is to be noted that, with the exception of the cases stated by STRANDBERG in 1920 and ALMKVIST in 1924/25, no closer data exist on any type determination of the causative agent in the abovementioned observations. STRANDBERG (S), however, grew a *Tr. violaceum* from infected hairs, and ALMKVIST (S) isolated from lesions on the scrotum, covered with crusts resembling scutula, an *A. Quinckeanum.*

In 1932 BRANDT (S) mentioned that during 1930 in the outpatient department for skin diseases in Stockholm out of 6,000 patients only 26, i.e. less than 0.5%, were seen with Dermatophytosis. In spite of the rarity of the disease the author, in 1931/32, had to deal on an estate near Stockholm with an epidemic of Trichophytosis, embracing 23 persons, and caused by *Tr. gypseum lacticolor.* The outbreak probably took its rise from an infected horse. It is of interest to notice that in the course of the epicemic the fungus showed an increase in virulence. Whereas in 1931 only superficial cases occurred, in 1932 suddenly cases of Kerion developed.

From Denmark a description of a case of Tinea favosa was given by LARSEN (D) in 1844. In a clinical report ENGELSTED (D), in 1864, mentioned the treatment of about 71 cases of Dermatophytosis, in 6 cases of which the infection had been contracted from cat. BORCH (D), in 1865, described these 6 cases more thoroughly, adding to them 19 further observations on the affection contracted from a kitten. To make sure about the identity of the lesions in the people and the kitten, BORCH (D) undertook an autoinoculation, and carried out cross-inoculations between men and different animals. Having also experimented with Favus, and finding a difference between this disease and Herpes tonsurans, the author concluded that these two diseases were caused by two different kinds of fungi, a conclusion that stood in poignant contrast to the opinions of the authorities of that time, represented by Tilbury Fox as well as Hebra, who believed the agent of mycotic skin affections to be just one and the same fungus.

In 1869 Bergh (D) described 3 cases of Onychomycosis. Further case descriptions of Dermatophytoses followed in annual clinical reports, and given by Bergh (D) (1869—1885), de Fontenay (D) (1875—1881), E. Pontoppidan (D) (1885), Rasch (D) (1898).

Bergh (D) in his report of 1879 points out that at the General Hospital of Copenhagen, a city of about 235,000 inhabitants, between 1866—1868 no more than 35 to 41 persons a year had been treated for Ringworm, and in 1898 Rasch (D) observed that between 1893 and 1897 at the out-patient reception at the Communal Hospital, Copenhagen, out of 8,462 patients 47 had suffered of Trichophytosis.

Ehlers (D), in 1892, while working in Paris, expressed his agreement with Djelalledin Mouktar's statement on the existence of a special type of mycotic lesions on the feet, presenting a further case of Trichophytia of the foot sole.

In Denmark, in 1893, Slomann (D) and Johansen (D) both reported on two cases of Kerion Celsi. In one of the cases reported by Slomann (D) the infection was probably contracted from a dog. In the cases reported by Johansen (D) it had, however, been transmitted from cows. Prip (D) reported in 1894 on an observation of megasporic Trichophytosis in two butcher's helpers.

However, no closer type determination of the causative agents was accomplished until 1907, when H. Bang (D) had to deal with a Microsporia epidemic, embracing 50 children in an orphan asylum. A culture here yielded a *Microsporum*, which fungus, according to a later explanation by Bang (D) (1910 c) proved to be *M. Audouini*.

In 1910 (a), Bang (D), working with Sabouraud in Paris, described a peculiar type of cutaneous Trichophytia, caused by an up to that time unknown *Dermatophyton*, *Trichophyton purpureum* Bang. This fungus, together with Castellani's *Epidermophyton rubrum*, described independently by the latter that same year, led to the acceptance of a new type of *Dermatophyton*, the *Trichophyton rubrum*.

That very same year, 1910 (b), Bang (D) also published a survey on the newer investigations on cutaneous fungus diseases, based on Sabouraud's papers, and demonstrated at the Meeting of the Northern Dermatological Society cultures of Dermatophytes encountered in Denmark, viz. *M. Audouini*, *M. equinum*, *Tr. violaceum*, *Tr. plicatile*, *Tr. gypseum asteroides*, *Tr. faviforme discoides*, *Tr. purpureum* (Bang). However, according to Sylvest (D) (1945), Bang (D) had in Denmark up to 1913 found in addition to these also *A. Schönleini*, *A. Quinckeanum*, *A. gypseum*, *M. pubescens*, *E. inguinale*, *Tr. gypseum radiolatum*, *Tr. gypseum granulosum*, and *Tr. regulare.*

Further there exist descriptions of a series of cases caused by the fungi just mentioned (HASLUND (D), 1911; L. NIELSEN (D), 1914; BANG & HASLUND (D), 1915; B. PONTOPPIDAN (D), 1915 1919; RASCH (D) 1915; 1916 (a, c), 1917, 1919, 1921, 1922, 1923, 1924 (a, b), 1925; BRUN PEDERSEN (D), 1916, 1917, 1918; FINSEN (D), 1925; GENNER (D), 1929, 1932; BOAS (D), 1930; EMANUEL (D), 1932; P. HANSEN (D), 1937; VOSBEIN (D), 1939).

BRUN PEDERSEN (D) in 1916 (c) had described two cases of Eczema marginatum caused by *Ep. inguinale* to occur in consequence of an application of wet dressings.

RASCH (D), who paid closer attention to the clinic of deep Trichophytosis, found that most of the cases in Denmark were caused by *Tr. faviforme discoides*, and in many cases, the origin of the infection could be traced back to cattle. The first observation of *Tr. rosaceum* in Denmark was made in 1919 by KISSMEYER (D), and in 1924 (b) RASCH (D) published further reports on the same fungus yielding also deep lesions.

In P. HANSEN'S (D) case (1937), a family consisting of a mother and 3 children had been infected with *A. Schönleini.*

Apart from EHLER'S (D) notation in 1892, more attention has been paid by Danish physicians to foot Mycosis. RASCH (D) in 1922, reported on the clinical features of interdigital Mycoses, but he did not cultivate the causative agent. Then LOMHOLT (D), in 1932 (a, b), demonstrated two cases of dysidrosiform foot lesions caused by *Ep. interdigitale*, and that same year (1932 e) reported on an investigation into the Eczema of feet, embracing 97 students at a boarding-school.

As appears from an in extenso publication in 1933, the majority of the examined students displayed eczematous skin changes of the feet, from which in 21 cases an *Ep. interdigitale*, in 3 cases an *Ep. inguinale*, and once an *A. gypseum* could be grown. Four of the cases were accompanied by nail changes yielding *Ep. interdigitale.* In one case the nail affections showed the characteristics of Leukonychia trichophytica.

Later, in 1935 and 1936, LOMHOLT (D) (1936) together with HALBERG (D), during a field examination of about 1,400 school children in Copenhagen, found that the occurrence of clinical symptoms of Epidermophytia pedis in children increased successively with their age, from being almost absent in elementary school pupils, and reaching a rate of about 50% in pupils at secondary schools.

In 1939 (a) HAXTHAUSEN (D) published a lecture on foot Mycoses, and in the same year BONNEVIE (D) issued the results of an investigation into the aetiology and pathogenesis of Eczema. He found the disorder in 16% to be due to Mycosis. Cultivation from mycotic lesions had yielded from feet *Ep. interdigitale*, *Ep. inguinale*, and once a *Microsporon*, resembling *M. Audouini*; from hands *A. gypseum*; from the inguinal region *Ep. inguinale*, *Tr. gypseum asteroides*, *Tr. crateriforme exsiccatum*; from the axillae *Ep. inguinale* and from nails *Ep. interdigitale.*

In Norway W. BOECK (N), in 1852, in his publications on the clinic of skin diseases, had given descriptions also on cases of Favus treated by him.

WINGE (N), in 1871, discussed a case of Onychomycosis. In 1887 KIAER (N) & C. BOECK (N) (1887 a) reported on observations of transmission of Herpes tonsurans from cattle, but also sheep, to man. HIORTH (N), in 1887, having examined 4,015 elementary school children in Kristiania, found among them Favus in 62 cases, and Herpes tonsurans in 33 cases. From 1878 to 1886 432 recruits had been exempted from military service on account of Favus. Of these recruits 24 came from towns, and 408 from the country. Besides those 2,410 persons had been treated in Norway for the same disease from 1868 to 1885.

From a report of the Medical Society, Kristiania, on infectious diseases in 1888, it appears that further observations on the transmission of Ringworm from cattle to man had been made. That same year by DAHL (N), and later, in 1911, by I. H. BIDENKAP (N) surveys on the occurrence of Favus and Trichophytosis in certain districts of the country were published. BIDENKAP (N), besides reporting on the transmission of Trichophytosis from cattle to man, also noticed that the infection may keep itself in old wooden buildings, and thus affect any fresh set of cattle placed there. He, however, did not investigate this way of transmission more closely.

In 1892 DANIELSSEN (N), in a monograph on plantparasitic skin diseases, had described a series of observations on Favus and Herpes tonsurans, in part of which he succeeded in transferring the infection to other patients, afflicted with leprosy. In persons already suffering from Favus the inoculation failed, or the site of inoculation, after a violent inflammatory reaction, healed by itself.

From Favus the author isolated *A. Schönleini*, and from Herpes tonsurans *Tr. Malmsteni*.

In an *A. Schönleini* culture DANIELSSEN (N) observed swollen terminal cells containing globular clumps. He reflected upon the possibility of these cells being asci, but he never saw the globules surrounded by any membrane, which would have permitted to accept them as ascospores.

In 1932 a trial lecture by GULDBERG (N) on Trichophytosis was published.

GJESSING & MOSSIGE (N) reported in 1936 on an observation of Epidermophytia in three brothers, one of whom displayed an atypical form of the disease, caused by *Ep. inguinale*, and, in 1937, HUBERT (N) demonstrated a case of pityriasiform Favus caused by *A. Schönleini.*

That same year MOSSIGE (N) (1937 a), reported on an observation of silver foxes acting as source of infection for Trichophytosis in man. The report was published together with DANBOLT (N) (1938 c, 1939 a, b). As the causative agent *Tr. gypseum asteroides* had been identified, and the authors pointed out that in a country where between 30,000 and 40,000 persons were daily engaged in silver fox raising, attention should be given to this animal as a source of fungous infection.

In 1938 MOSSIGE (N), who had got his mycological training with MUSKATBLIT in New York, reported on the first findings of *Ep. rubrum* (CASTELLANI-BANG) in Norway, the fungi originating from three cases of foot and hand Mycoses. The author was inclined to regard the different strains presented in literature as varieties of the same species only.

In 1940 MOSSIGE (N) presented a survey on Epidermophytia pedis and its problems, reporting simultaneously on the occurrence of the disease in Norway, where it seemed to be rather widespread. The author found it in the half of 50 persons, chosen at random for examination. He also noticed that susceptibility to foot Mycosis seems to differ highly in different individuals.

From Finland ENVALD (F), in 1890, reported on the occurrence of Herpes tonsurans, and, in 1891, on Onychomycosis caused by Trichophyton and Favus.

Atypical and rare cases. In 1844 F. T. BERG (S) observed Favus in an infant, displaying scutula on the arm only. H. BANG (D) reported in 1910 (e) on a case of simultaneous occurrence of Favus and Trichophytosis in the capillitium. Dermatophytosis in infants was also observed by RASCH (D) in 1913 (a) and 1925 (a). The youngest of the children had shown signs of the disease at 6 days' age (1913 a). In 1923 RASCH (D) reported on a case of Favus in the beard region caused by *A. Schönleini*, and in 1924 (a) on a case of Microsporia caused by *M. Audouini* in the hair of a 54-year-old man. STRANDBERG (S) had in 1920 seen a case of Trichophytia capillitii caused by *Tr. violaceum*, clinically resembling a Microsporia. In 1937 GJESSING & MOSSIGE (N) described a case of an extensive diffuse scaling of the scalp and the face caused by *Ep. inguinale*, and in the same year HÜBERT (N) demonstrated a case of pityriasiform Favus caused by *A. Schönleini*, yielding only scales, but lacking scutula or atrophy.

Transference from animals. Besides the cases, of Dermatophytosis contracted from animals already mentioned, there also exist a series of observations by veterinarians on the transmission of the disease from cattle (LINDQUIST (S), 1867, ENGSTRÖM (S), 1916) or horses (BENGTSON (S), 1923, HERMANSSON (S), 1936, H. FRIIS (D), 1937) to man.

In the case described by HERMANSSON (S) the infection had been transmitted to more than 12 persons from a horse, as it had passed from hand to hand by repeatedly being sold.

Occupational diseases. In 1929 VENNERHOLM (S), in a review on occupational diseases in veterinarians, mentioned Favus and Trichophytosis, and from a survey by THOMSEN (D) (1937) into the occupational diseases in Danish veterinary surgeons, completed during 1935, it appeared that out of 960 veterinarians 159 had suffered of Ringworm, many of them several times.

In 1920 B. PONTOPPIDAN (D) had demonstrated a case of "occupational" Dermatophytosis in a 9-year-old boy. The boy, who had a Trichophytosis of the right hand, used to lather customers in a barber's shop, which he had been doing for two years.

Allergy. Dermatophytids. Inga SAEVES (N), working with JADASSOHN in Bern, published in 1915 the results of an extensive experimental research into Trichophytids. She showed that secondary eruptions in Guinea pigs can be produced haematogenously. She also drew attention to the existence of a corymbiform type of Trichophytids. BRUUSGAARD (N) (1919, 1920, 1922) confirmed in 1920 that the haematogenous spreading of Trichophytids, at least in part of the cases, holds good also for man. He demonstrated histologically fungous elements with secondary nodules not only in the pars reticularis cutis, but also in the capillaries within the papillae, being able at the same time to grow an asteroides-like *Tr. gypseum* from the secondary efflorescenses, as well as from the primary lesions.

Since 1915 also a series of descriptions on the different kinds of clinical manifestations of the Dermatophytids have been presented by RASCH (D) (1915, 1916 a,c, 1917, 1919) and his school (BRUN PEDERSEN (D), 1916 a,b, 1917, 1918, B. PONTOPPIDAN (D), 1919, HAXTHAUSEN (D), 1920, 1929, H. BANG (D), 1923, LOMHOLT (D), 1932 a,b,d, KISSMEYER (D), 1934, BONNEVIE (D), 1939 a, VOSBEIN (D), 1939). In addition to the usual papular lichenoid eruptions, various other types of Ids were observed. Thus pure vesicular, morbilliform, as well as scarlatiniform ones, and eruptions resembling a multiform erythema, or scaling, reminding of Psoriasis. Also pigmentation and Alopecia in minute patches in consequence of Ids were mentioned. In a case observed by HAXTHAUSEN (D) (1929) the Ids were located in areas with vascular changes due to Pernio. In KISSMEYER'S case and in one of the cases mentioned by BONNEVIE (D) the Ids were provoked by an intracutaneous Trichophytin injection.

The bulk of the cases developing Ids proved to be caused by *Tr. faviforme*. Only VOSBEIN'S (D) case, with an Erythema multiforme-like eruption, yielded in cultures *A. gypseum*. The dysidrosiform lesions, however, accompanying foot Mycoses, were due to *Ep. interdigitale*, as pointed out by LOMHOLT (D) (1932 a,b).

In Sweden an occurrence of Trichophytids has been mentioned by STRANDBERG (S) (1931), and HELLERSTRÖM (S) (1934).

Trichophytin reaction. MARCUSSEN (D) had in 1937 and 1940 studied the immediate reaction with intracutaneous Trichophytin injections in about 2,000 eczematous persons. In 1937 (a) he had, moreover, examined 8 cases of urticarial reactions to Trichophytin more thoroughly.

The author concluded that the immediate reaction depends on the mycotic infection as well as on an allergic disposition of the individual. The immediate and the delayed Trichophytin reactions consume the same antigene component, but an immediate reaction can be called forth also by primary toxicity of the Trichophytin, or hypersensibility towards certain components of the culture mediums on the part of the person tested, and need therefore not be of absolute diagnostical value.

In 1939(a) BONNEVIE (D) stated that Epidermophytia is always followed by a hypersensibility against Trichophytin. Positive patch tests, however, are only encountered in persons with a certain degree of hypersensibility, viz. in those who react to an intracutaneous injection of 0.1 ccm of Trichophytin with at least 20 mm.

Therapy. In Scandinavia the treatment of Dermatophytoses has on the whole followed the pattern carried out elsewhere. With superficial lesions the common antimycotics have been used, such as Iodine, Sulphur, Mercury compounds, Salicylic acid, Benzoic acid, Resorcine, Chrysarobine, Anthrarobine, Carbolfuchsine, Genriana violet, Brilliant green etc. (ENGELSTED (D), 1879; E. PONTOPPIDAN (D), 1885 b, 1890, 1891, 1896; DANIELSSEN (N), 1892; RASCH (D), 1905, 1909, 1927; AFZELIUS (D), 1915, 1925; LOMHOLT (D), 1934, 1938 b, 1939). RASCH (D), however, seems to have avoided Mercury compounds. Earlier attempts had also been made with Camphor (SUNDEWALL (S), 1871), Phenolic acid (NORDENSTRÖM (S), 1872), and Boric acid (FOLLUM (N), 1888).

With deeper infections, the so-called Dermatophytosis profunda, lenients were applied, and in resistent cases epilation of the hair, and also the beard, was effectuated (W. BOECK (N), 1852; DANIELSSEN (N), 1892; RASCH (D), 1905, 1909, 1927; LOMHOLT (D), 1939).

In older times epilation had been carried out with a pitch cap. But, as that procedure was too heroic, W. BOECK (N) had in 1852 recommended replacing the pitch cap with some kind of plaster. Later on they took to manual epilation with pinchers. But also electrolysis of the infected hair roots was practised (RASCH (D), 1905, 1909, 1927).

At the beginning of this century X-rays were introduced for epilation (SEDERHOLM (S), 1903; SJÖGREN (S), 1904; REYN (D), 1905, 1909; JERSILD (D), 1909), SABOURAUD'S method becoming routine for the treatment of the capillitium (RASCH (D), 1909). Nevertheless, it was not without a dispute by letter, in 1907/08, with FISCHER (D) (1907, 1908) against, and H. BANG (D) (1907), RASCH (D) (1907 b, 1908), as well as SABOURAUD himself (1907, 1908) for it, that the question finally became settled in favour of the general use of the method.

In case the X-rays failed to depilate completely, L. NIELSEN (D), in 1914, recommended the use of an Ichthyol-cotton cap to accomplish the task.

Of incidences in consequence of X-ray application, RASCH (D), in 1907 (a), reported on a case of Alopecia and mental disturbance, viz. Pyromania, following X-ray treatment of Microsporia.

O. JERSILD (D), in 1925, demonstrated two siblings with naevi in the capillitium, suffering from Trichophytosis, in whom the hair did not loosen within the limits of the naevi, in spite of X-ray application.

Besides the conventional therapeutical methods W. BOECK (N), in 1852 (a), had tried the scarification method with Favus, but without success. WELANDER (S), in 1900, tried heat-treatment with Herpes tonsurans capillitii. He attempted to kill the fungus by using a constant flow of water, heated to 45° C, and led through pipes, which in their turn were adapted for an application as a cap. Although promising in certain respects, the method, on the whole, proved too awkward for a more widespread use (SEDERHOLM (S), 1903).

In 1921 H. BANG (D), and in 1925 (b) RASCH (D) reported on a successful treatment of Trichophytia, caused by *Tr. rosaceum*, resp. *Tr. faviforme album*, by intravenous, resp. internal application of Potassium iodide.

In 1934 O. JERSILD (D) proposed Copper-iontophoresis treatment for Dermatophytosis of the extremities (O. JERSILD (D), 1934; O. JERSILD & PLESNER (D), 1936, 1939, 1940; KISSMEYER (D), 1938), and LOMHOLT (D), that same year, introduced a new antimycotic Ethylparaoxybenzoate, or Mycocen, for the treatment of Dermatophytosis (LOMHOLT (D), 1934 a, b. 1938; BONNEVIE (D), 1935, HAXTHAUSEN (D), 1936; H. BANG (D), 1937).

With Copper-iontophoresis treatment, according to O. JERSILD & PLESNER (D) (1939, 1940), a cure was achieved in 87% of 79 cases. However, no after-check was made by the authors. Sharing KISSMEYER'S (D) (1939) opinion, it would have been difficult to differentiate between relapses and re-infection. As to the effect of Mycocten, HAXTHAUSEN (D), in 1936 reported on the cure of Favus in two sibling, and H. BANG (D), in 1937, had achieved a cure in 5 cases of Microsporia capillitii, caused by *M. pubescens*, a variety of *M. lanosum*. Nevertheless, BONNEVIE (D), in 1939 (b) observed on one occasion a sensibilization by the compound.

During the earlier period nail infection had been treated principally with Potassium iodide solution, applied as dressings, but also with Benzoic, as well as Salicylic acid, employed in unguents (RASCH (D), 1905, 1909, 1927). Later ablatio of the nail plate was recommended (LOMHOLT (D), 1939).

The prophylaxis of foot Mycosis has been amply discussed by LOMHOLT (D) (1934 c, 1939, 1940), but also by MOSSIGE (N) (1940). The authors recommended bath establishments to be built of hard material, not to be soaked by moisture, and for the desinfection of the establishments solutions of 5—10 % Sodium hypochloride, or 10—20% Sodium hyposulphate were recommended. In regard to individual prophylaxis concerning the desinfections of the feet, solutions of different kinds of antimycotics were prescribed, and LOMHOLT (D) also proposed the wearing of rubber toe protectors. The authors advised stockings to be desinfected in Methyl alcohol, and for foot wear the same liquid, or a solution of Formaldehyde was recommended. With Formaldehyde however, people had to be careful in respect to possible sensibilization.

BLASTOMYCOSIS, INCLUDING CANDIDIASIS

After F. T. BERG'S (S) and HANNOVER'S (D) studies in the early 1840—ies, as mentioned in the introduction, into the occurrence of yeast-like fungi in the oral cavity and the adjacent parts of the respiratory tract, as well as in the oesophagus, in 1869 a report was published by REISZ (D) on the finding in a post-mortem of *Oidium albicans* on the superficial layers of the mucous membranes of the ventricle.

In 1890 BENTZEN (N) declared on hand of two clinical observations that Thrush may occur idiopathically, and without serious consequences to the affected person, in adults suffering of only slight acute disorders. This statement by BENTZEN (N) was, in 1894, confirmed by K. GRØN (N) on the basis of three more, similar observations.

In 1899 LUNDSGAARD (N) reported on a case of Hypopyonkeratitis, yielding pure cultures of yeast. BERGHOLM (F), in 1902, examining the vaginal secretion of pregnant women, found on the basis of 40 cases, the smaller, aerob part of the flora to consist, besides of a bacillus, of yeasts. In 1915 NORRLIN (S), and in 1926 v. WENDT (F) cultivated from the contents of the abdominal cave in cases of peritonitis in consequence of perforated ulcers of the digestive tract, besides microbes and an aerob *Streptothrix*, also yeasts. Moreover, v. WENDT (F) found the flora on these occasions to be a reduced one of that of the oral cavity.

MÜLLERN-ASPEGREN (S), in 1921, reported on a case of Cystitis accompanied by vigorous gas formation in the bladder. The urine was of a high sugar concentration, and microscopically displayed abundant yeast cells. The condition improved after local irrigations with Boric acid solution.

In 1922 RASCH (D) described the clinical features of a series of cutaneous yeast Mycoses, simultaneously pointing out the difficulties in making a proper diagnosis in the case of such disorders. Whereas some authors spoke only of ,,yeasts'', others immediately identified the causative organisms with Thrush. The question was the more closely studied by H. FINSEN (D), who was in that matter helped by the Carlsberg Laboratory, Denmark, which since the days of Emil Chr. HANSEN (D) had been a centre for the study of yeasts.

Finsen's (D) report appeared in 1924. The author had succeeded in cultivating yeast-like fungi from 27 persons, suffering of superficial mycotic lesions, and twice from samples of interdigital scrapings from 40 healthy individuals. He, called the fungi *Torula* on account of lack of spore formation. The bulk of them fermented glucose and maltose with gas production, whereas saccharose and lactose did not give any gas.

Later, in 1930, Finsen (D), isolating different kinds of *Monilias*, from skin affections of infants stated that the lesions need not necessarily originate from a contamination from the oral cavity.

In 1928 Haxthausen (D) presented a case of Blastomycosis with ulcerative nodules on the left cheek, and in 1932 (b) and 1935 two cases of generalized Moniliasis displaying wide-spread lesions over the tegument with an involvement of the finger nails and the mucous membranes of the oral cavity. Cultures from these cases yielded a yeast, and a *Monilia* species respectively. The case of Blastomycosis, and one of the Moniliasis improved after treatment with Potassium iodide per os.

Earlier, in 1913 (b), Rasch (D) had got into a pitfall, assuming in a case of chronic verrucous ulcerations on the back of a hand, with cultures yielding three different kinds of yeasts (Rasch & Kissmeyer (D), 1914), the affection to be a Blastomycosis. In 1916 (b), he, however, was on the basis of histopathological findings able to announce that the case had finally been recognized as skin Tuberculosis.

In 1933 Ottander (S), and in 1934 Schnell (S), respectively reported on a case of Pneumomycosis by *Oidium albicans*, and a case of mucopurulent Bronchitis by an *Oidium* species. In both cases the fungi were looked upon by the authors as the causative agents of the disorders. Ottander's (S) case improved after Potassium iodide application. Schnell (S), however, did not dare to use Iodin therapy on account of an old Tuberculosis with the patient, but contented himself with the treatment of the Diabetes the patient also displayed.

Danbolt (N), in 1938 (a), described a case of fatal progressive Moniliasis of the skin, caused by *Candida stellatoidea*. Histologically yeast like organisms were present in subcutaneous layers. On blood-agar the strain yielded star-shaped colonies, and in the fermentation test it produced gas with glucose and maltose, and coagulated milk. The same year Møller (D) reported from the Farøe Islands on a case of Diabetes mellitus, accompanied by an infection of the urinary tract with a yeast-like fungus, which fermented glucose, galactose, saccharose, and lactose.

Kristenson (S), in 1940, checking the question of the occurrence of a yeast-like fungus in connection with tuberculous affections, encountered the fungus twice in 104 patients with Tuberculosis, and 9 times in 174 patients suffering from different other ailments. Thus no correlation with Tuberculosis could be established, but the individuals yielding positive fungus findings were found to be in a rather poor physical condition.

CRYPTOCOCCOSIS

Cryptococcus neoformans, as already mentioned, had been used in 1900 by STECKSÉN (S), and in 1902 by V. P. H. JENSEN (D) for experimental studies into the aetiology and the histogenesis of neoplasmata. Further V. P. H. JENSEN (D), in 1903, showed in his thesis that a series of yeast-like fungi studied by him, resembled in all respects *Saccharomyces neoformans* SANFELICE. Nevertheless, he still set apart the yeast-like fungi isolated by BUSSE and by CURTIS.

All the strains used for the above-mentioned studies had, however, been of foreign origin, and it was not until 1923 that VOSS (N) announced the first case of Cryptococcosis met with in Scandinavia. He described it as a generalized Blastomycosis, caused by *Saccharomyces hominis* VUILLEMIN, running under the picture of a Meningitis of fatal outcome.

PITYROSPORUM OVALE

Microscopic slides of MALASSEZ' spores, or *Saccharomyces sphaericus* and *ovalis*, BIZZOZERO, were demonstrated by C. BOECK (N) in 1885 and 1887 (b). In 1888 the same author claimed to have been successful in cultivating the fungus on potato medium.

SPOROTRICHOSIS

Literature surveys on Sporotrichosis have been published by K. GRØN (N), in 1909; JENSEN C. E. and KIØRBOE (D), in 1910; and BRUUSGAARD (N) in 1912, but the disease itself was encountered in Scandinavia only in 1912 by WIDRÖE (N) in Norway. In 1914 C. BOECK (N), and in 1934 VOSS (N) reported on further cases. In Denmark Sporotrichosis had been observed only once, viz. in 1918 by RASCH (D).

All the cases, except that of VOSS (N), which was a pulmonary one, were of a cutaneous type, and in each case the diagnosis had been verified by cultures. All the cases improved by the conventional therapy with Potassium iodide.

PITYRIASIS VERSICOLOR

Reports on taking care of persons suffering from Pityriasis versicolor we find in clinical records by W. BOECK (N) as early as 1852.

In 1863 BERGH (D) announced an observation of microscopically verified Pityriasis versicolor in a 2 months' old infant, which is probably the first report on the disease occurring in so young an individual. Attention was anew drawn to that observation by the author in 1894, in connection with an observation by NIELSEN (D), that same year, of the disease in a 9 months' old child.

PETERSEN (D), in 1914, demonstrated two cases of Pityriasis versicolor, where the lesions on sun-burnt skin looked white.

For the treatment of Pityriasis versicolor the common remedies such as Sulphur, Iodine, Salicylic acid, or Resorcin have been used (RASCH (D), 1905, 1909, 1927; LOMHOLT (D), 1939). Later also Ultraviolet rays were recommended (LOMHOLT (D), 1939). In connection with the treatment, also the necessity for a desinfection of the clothes was pointed out.

PIEDRA

SVITZER (D), in 1845, had observed a case of white Piedra in Denmark, and published a closer description of the affection. Unfortunately the author did not recognize the mycotic nature of the disorder, interpreting the nodules on the hairs as conglomerations of epithelical elements, and therefore simply called the affection an „uncommon disease of the hair".

MOULDS

Fusarium. From 1852 there exists a report by LEVIN, WAHLBERG & HÖK (S) on wide-spread incidents of intoxication among the population of Southern Sweden, caused by the consumption of rye, spoilt by *Fusarium heterosporium*.

Aspergilli. The first man-pathogen Aspergilli in Scandinavia were discovered in Otomycosis, in 1871. NICOLL (N) cultivated from one case an *A. glaucus*, and LILJENROTH (S) (1871, 1872) isolated from 6 cases *Aspergillus flavescens*, which fungus, however, according to his drawings, resembled an *A. fumigatus*. Histologically the fungi formed a mycelial network with fully developed conidia bearing heads in the layer of epithelial débris in the auditory canal.

In 1877 and in 1883 two further cases were reported by af SCHULTÉN (F) and v. ESSEN (F). Both cases yielded an *A. flavescens*, probably the same fungus as encountered by LILJENROTH (S).

JOHAN-OLSEN (N), in 1886 (a), isolated an *A. niger* from pustular skin lesions arisen in consequence of LISTER's bandage, and the same year (1886 b), he, together with GADE (N) described and proved the pathogenicity of *A. subfuscus*.

Aspergillosis of the lungs in man, caused by *A. fumigatus*, was described by BERGMAN (S), 1929; BERGMAN & HENSCHEN (S), 1930; BERGMAN, DANIELSON (S), 1933; and DANIELSON (S), 1933, the diagnosis having been made in post-mortems, carried out by HENSCHEN (S), and SCHNELL (S). The authors were in agreement with v. HELLENS (F), who, in 1902 and 1905, studying Pneumomycosis in sheep caused by the same fungus, had concluded that

A. fumigatus should not be made responsible for any cavern production in the lungs.

In 1931 KANERVA (F) described a case of Keratitis aspergillina, caused by *A. fumigatus*. Instead of operating on the eye, as recommended in literature, the author had treated it succesfully with Xeroform powder and a 2% Noviform-Atropin unguent.

Penicillia. The first description of the Penicillin-yielding *Penicillium notatum* has been given by WESTLING (N) in 1911. In 1920 NORGAARD (D) found Penicillia to be the most often occurring moulds among the contaminants isolated from samples of spoilt morphin solution, collected at different pharmacies.

In 1926 (b) GUNDERSEN (N) reported on the cultivation of a *Penicillium* species from urine in a case of dysuria with a patient suffering from an obscure lung affection. In the urine the fungous elements appeared, besides ramified hyphae, partly as cylindrical bodies composed of spores. The report gave rise to some critical remarks as to the mode of determining the fungus from (JOHAN-OLSON) SOPP (N), 1926. But this criticism was calmed down by a declaration by GUNDERSEN (N) (1926 c), who stated that he had actually made the diagnosis on the basis of the fungous morphology in the aerial mycelium.

Scopulariopsis. In 1925 RASCH (D) presented a case, and in 1933 and 1939 (a) BONNEVIE (D) mentioned five more cases of Onychomycosis caused by *Scopulariopsis brevicaulis*.

Mucor. HAFSTRÖM (S), in 1930 and 1933, reported on a case of lung tumor, in which, in the course of the illness, there had repeatedly been isolated a *Mucor*, presumably *M. rhizopodiformis*. The fungus might, according to the author, however, have only been a secondary invader to the patient's lung cancer, which latter diagnosis was made with greater exactness only at the post-mortem.

In 1939 LEHMANN & H. E. NIELSEN (D), and in 1940 H. O. BANG (D) employed for clinical Thiamin analysis of the blood SCHOPFER'S mould method with *Phycomyces blaksleeanus*. The authors were quite satisfied with the method.

AIR-BORNE FUNGI

Data on air-borne fungi from Scandinavia have been published as early as 1879, 1882, and 1887 by Emil Chr. HANSEN (D), in 1887 by SELANDER (S), and in 1908/09 by ROSTRUP (D).

Both HANSEN (D) and ROSTRUP (D), the latter of whom had accomplished his investigation in 1903/04, found among the moulds in Denmark the most frequent to occur to be *Cladosporium* or *Hermodendron* species, *Penicillium glaucum*, *Dematiatium pullulans*, and *Botrytis cinerea*. Then, according to HANSEN (D), followed *Mucor stolonifer* (*Rhizopus nigricans*), *Mucor racemosus*, and an *Arthrobotrys* species. ROSTRUP (D), however, did not encounter these species. According to him there followed *Trichocladium asperum*, *Mucor spinosus*, and a *Macrosporum* species, which fungi HANSEN (D), in his turn, had seen only very sparsely. Of other kinds there had occurred species of *Aspergillus, Alternaria, Fusarium, Verticillium, Phoma, Stachybotris, Oidium* etc. But also yeasts were encountered, white, red, as well as black varieties.

HANSEN (D) also pointed out a seasonal variation in the presence of different kinds of fungi in the air, in winter the moulds being more resistant against cold. ROSTRUP (D) furthermore observed that the count of the germs outdoors was 6 to 7 times as high as indoors. Whereas outdoors *Cladosporium* or *Hormodendron* predominated, indoors *Penicillium* was the most frequent one to occur.

In 1879 HANSEN (D) had studied the air content of fungi with regard to the deterioration problem in the brewery industry, and ROSTRUP (D) had done it of purely academical interest. In 1882 and 1887, however, HANSEN (D) analyzing his findings, pointed out their possible interest for hygienists, too.

SELANDER (S), in 1887, examining the air in a cantonment, found it, inter alia, to cantain scantily *Penicillium glaucum, Oidium lactis, Botrytis cinerea, Aspergillus niger*, and a red yeast, called *Saccharomyces glutinis.*

Some remarks on sensitivity to moulds we find only in 1934, in a survey on the allergical type of Asthma bronchiale, published by ERIKSSON (F).

BASIDIOMYCETES

HELLERSTRÖM (S), in 1940 described two instances of cutaneous sensitization to *Boletus edulis* BULL, *Boletus luteus* L, *Lactarius deliciosus* (L), and *Clavaria flava* SCHAEFF, in consequence of handling raw edible mushrooms. In one instance, hypersensibility was specific only for one kind of mushroom of the same family, *Boletus*. In the other case hypersensitivity was shown also after eating the mushrooms, fried. Boiling seemed to destroy the sensitizing agents.

LINGUA NIGRA

The explanations of the findings of fungi in Lingua nigra or Black tongue has, like all over the world, caused trouble also for the physicians in Scandinavia.

SELL (D), in 1879 and 1885, as well as later, in 1908, BLEGVALD (D), studying the occurrence of fungous elements on the affected tongue, came to the conclusion that the fungi there should be looked upon only as secondary invaders to previously hypertrophized tongue filaments.

Although BÖCHER (D), in 1887, on account of microscopical examination of two cases, and SCHMIEGELOW (D), in 1896, because of the isolation of dark-coloured moulds from two further cases, had sided with the parasitic theory, the deductions of these authors seemed to be erroneous (BLEGVALD (D), 1908).

ACTINOMYCETALES

In 1890 Almquist (S), who had been instructed about the cultivation techniques by Johan-Olsen (N), published a study on three aerobic *Streptothrix* strains, yielding aerial mycelium that broke up into spores. The author, however, regarded these organisms as pleomorphic bacteria.

The first to cultivate Actinomycetes in Finland, according to v. Bonsdorff (F) and to Äyräpää (F), both in 1894, had been Cygnaeus (F).

Harbitz (N), as a result of his bacteriological studies, from 1898 to 1911, into the aetiological agent of Actinomycosis, could confirm Wolff-Israel's views about the anaerob nature of the organism.

In 1924 Thjøtta & Gundersen (N) reported on the isolation of a *Streptothrix* from the blood of a case of acute rheumatism. The authors could, however, not establish any pathogenicity for the strain. Also another fungus strain, described in 1913 by Sopp (N) in connection with a case of human Actinomycosis, seems, on account of the occurrence of conidia in it, to have been a *Streptothrix*.

Naeslund (S) settled in 1931 an old controversy, showing definitely a difference to exist between the pathogenicity of the aerob and the anaerob types of Actinomycetes. He also drew attention to the facultative anaerob properties of certain strains, resembling in other respects the strictly anaerob ones.

However, most of the basic research into Actinomycetales in Scandinavia has been done in Denmark. Ørskov (D) in 1923 and H. L. Jensen (D) in 1931, e.g. published each a classification for these organisms.

Ørskov (D) divided the Actinomycetes into three groups: I. Cohnistreptothrix — organisms which form an aerial mycelium dividing into spore-like bodies. II. Actinomyces — organisms with mycelium divided into fragments of irregular shape, rod-like, or granules. This group was further divided into two sub-groups, II a and IIb, the organisms within the latter displaying a tendency to adopt angular growth. III. Micromonospora — organisms with spore-like bodies borne singly on the tips of short branches of the vegetative hyphae.

The designation ,,Micromonospora'' is still in use as a genous name for spore-bearing organisms, comprising many thermophilic strains.

Jensen (D) divided the organisms into two main groups: I. Proactinomycetaceae — organisms without any spore formation, coining here a special name, ,,Proactinomyces'', for a sub-group, nowadays known as *Nocardia*. II. Actinomycetaceae — organisms with spore production.

Naeslund (S), in 1940, agreed with the point of view that the Actinomycetes should be classified in regard to their morphological characteristics.

Although the classification of Ørskov (D) and Jensen (D) were not generally accepted, still, their attemps have added much to our comprehension as to the definition and the systematization of the Actinomycetales.

In 1930 Holm (D) had shown experimentally that 9 strains of anaerobic Actinomycetes, obtained from Actinomycosis in man, with regard to their agglutination and fermentative properties fell into two distinct groups. v. Magnus (D), in 1931, observed an *Actinomyces*, belonging to Ørskov's group IIb, to yield a slime capsule under certain growth conditions. And in 1938 Ørskov himself saw motility in some strains of group II.

Actinomycosis in Scandinavia was in 1883 reported from Denmark by R. Jensen (D) to occur in cattle. Odenius (S) mentioned, that same year, an observation of actinomycotic lesions in the liver of a turkey. According to Ljunggren (S) (1895), Odenius had actually made this observation as far back as 1874, but had been reluctant to give any explanation to his findings before he had learned about the discovery of a new disease, the Actinomycosis.

In 1883 also B. Bang (D) published his exhaustive treatise into Actinomycosis, and in 1884 Gade (N) published a literature survey on the subject.

Nevertheless, it was not until 1887 that the first human cases were observed in Denmark by Rovsing (D) and by F. Kaarsberg (D), and in Norway by Conradi (N) and A. holst (N). The latter answered for the post-mortem records of the case, which he later, in 1888, even completed by a further publication. From Sweden the first cases were reported in 1890 by Lennander (S), and J. Berg (S), and from Finland in 1892 by v. Bonsdorff (F). v. Bonsdorff's report, embracing two cases, was followed that same year by a post-mortem record of one of them by Homén (F).

After these first case reports literature reviews on Actinomycosis in man were also published in Denmark, in 1889, by Rovsing (D), and in Sweden, in 1891, by Åkerman (S).

Further casuistics in human Actinomycosis followed from Denmark by Ammentorp (D), 1888 and 1893; Tscherning (D), 1893; Rasch (D), 1898; J. Friis (D), 1898; Hertz (D), 1909 and 1935; Frölich (D), 1910; Lodberg (D), 1911; Videbach (D), 1914; J. Kaarsberg (D), 1914; Nordentoft (D), 1914 and 1933; Riising (D), 1921; Christensen & Eiken (D), 1924; Eiken (D), 1926; and onward, Gelbjerg-Hansen (D), 1930; Lindegaard Nielsen (D), 1931; Rischel (D), 1931; Søndegaard (D), 1935; Bruun (D), 1938; Andersen (D), 1939, From Norway by Ramm (N), with a post-mortem record by Kjerschov (N), 1892; Laache (N), 1892 and 1906; Friele & Hartwig (N), 1894; P. Holst (N), 1895 and 1897; Jervell (N), 1895; Cappelen (N), 1896; Herlofsen (N), 1897; Harbitz (N), 1898, 1909 and 1910; Harbitz & Backer Grøndahl (N), 1910; Thesen (N), 1909; Scheen (N), 1912; Holland (N), 1913; Heyerdahl (N), 1914; K. Nicolaysen

(N), 1918; N. A. NICOLAYSEN (N), 1920; N. A. NICOLAYSEN & STOLTENBERG (N), 1923; MOTZFELDT & THJØTTA (N), 1924; SERCK-HANSEN & SWENSSON, 1927 (N); HØYGÅRD (N), 1935; DANBOLT (N), 1940. From Finland by J. W. RUNEBERG (F), 1892; AF SCHULTÉN (F), 1893 and 1897; KROGIUS (F), with a post-mortem record by HOMÉN (F), 1893; ÄYRÄPÄÄ (F), 1893; v. BONSDORFF (F), 1894 and 1901; AF FORSELLES (F), 1900; BROTHERUS (F), 1913; HJELMMAN (F), 1914; B. RUNEBERG (F), 1914; LANGENSKIÖLD (F), 1916 and 1926; BROFELDT (F), 1921 and 1926; A. KROGIUS (F), 1926; LINDGREN (F), 1931. From Sweden by SÖDERBAUM (S), 1893; KÖSTER (S), 1893 and 1894; EKEHORN (S), 1895, 1896 (a), and 1900; LJUNGGREN (S), 1895; BORELIUS (S), 1896 and 1903; ELIASSON (S), 1895 and 1896; BÄÄRENHIELM (S). 1900; DALÉN (S), 1902; SÖDERLUND (S), 1908, and onwards up to 1927; EDBERG (S), 1916; TURESSON (S), 1916; HALLDÉN (S), 1917; H. SUNDBERG (S), 1919; NORLÉN (S), 1925; MANFRED (S), 1926; BERGENHEM (S), 1929; AHLSTRÖM (S), 1930; S. BERG (S), 1930; AHLTORP (S), 1933; KNUTSON (S), 1933; v. SYDOW (S), 1936.

Data on the frequency of the disease in regard to Finland have been given by v. BONSDORFF (F), 1894, and BROFELDT (F), 1921. and 1926; in regard to Sweden by HELLSTRÖM (S), 1895, BORELIUS (S), 1896, and EKEHORN (S), 1896; in regard to Norway by HARBITZ & BACKER GRØNDAHL (N), 1910. In most of these papers it has also been indicated how the geographical occurrence of Actinomycosis in man and animal followed each other.

In Norway HARBITZ (N) was the first to carry out some research work into Actinomycosis (1898), and the paper, published in 1910, together with BACKER GRØNDAHL (N), and based on 83 cases, included also the pathological anatomy and bacteriology of the disease. In addition to the paper by HARBITZ & BACKER GRØNDAHL (1910), HAGEM (N) had tried to elucidate the question of the occurrence of pathogenic Actinomycetes in nature. However, he did not encounter any anaerob organisms either in the air, or on plants, or in the earth, and he succeeded in isolating an anaerob *Actinomyces* only from a living being, viz. an infected calf.

In Denmark a more extensive research in the clinical field has been accomplished from 1924 (CHRISTENSEN & EIKEN (D)), and 1926 onward by EIKEN (D), who dealt with the problems of the diagnosis and the X-ray therapy of the disease.

In 1930 EIKEN (D) stated, as also BERGENHEM (S) had done in 1929, that findings of granules need not be pathognomical for Actinomycosis, and that microscopically gram positive threads are of far greater importance for the diagnosis than gram negative clubs.

In Sweden, during a period from 1909 to 1927, SÖDERLUND (S), partly in collaboration with NAESLUND (S), who in connection with his research into ray-fungi performed the greater part of the experimental work, elucidated the aetiological role of the Actinomycetes in the formation of salivary calculi.

According to NAESLUND (S) the Actinomycetes have a physical importance for the formation of tartar. Furthermore, the precipitation of lime salts is a consequence of the biochemical processes of these organisms. These views of NAESLUND'S (S) about the role of Actinomycetes in the formation of concretions in the oral cavity, besides *Leptotrix*, were maintained also by GRYTHE (N) (1933, 1938), who, moreover, in 1939, issued a survey on the occurrence of Actinomycetes in nature and warm-blooded animals. Also MODÉN (S), who had in 1935 found Actinomyces-like granules in 49% of 284 cases of chronic tonsillitis, showed in 1938 by staining the granules according to GRAM, as well as by cultivation that those granules, like tartar in the oral cavity, or calculi in the salivary glands, were built up by Actinomyces-like microorganisms.

Earlier, in 1890 and in 1891, the role of the oral cavity as a source of infection for Actinomycoses had been pointed out by HADERUP (D) in his lectures. It was, however, not until later that experimental proof was established for this statement. In 1925 (a) NAESLUND (S), and in 1936 v. MAGNUS (D) showed that Actinomycetes of the more or less anaerobic type constituted part of the normal mouth flora. Moreover, in 1932, BJERRUM & HANSEN (D), had, in collaboration with HOLM (D), succeeded in isolating anaerobic Actinomycetes from noncharacteristic acute inflammatory lesions in the oral cavity, as well as the cervical region. Furthermore, LØVDAL (N), in 1937, reported on a case of Actinomycosis after a tooth extraction.

The treatment of Actinomycosis in Scandinavia and Finland was in the beginning surgical, but soon also Potassium iodide was applied (MALM (N),1893; NICOLAYSEN (N), 1893, SCHOU (D), 1895, ELIASSON (S), 1896, F. GRØN (N),1915, N. A. NICOLAYSEN (N), 1920, TAKALA (F), 1922, HAXTHAUSEN (D),1932 a). In 1939 even Iodine-iontophoresis was tried (TAWAST-RANCKEN (F)).

Attempts with Actinomyces vaccine were made by HARBITZ & BACKER GRØNDAHL (N),1910, BACKER GRØNDAHL (N), 1918, and WISSING (D), 1928. But the results proved too controversial to draw any conslusions as to its therapeutical value.

In 1914 HEYERDAHL (N) introduced Radium therapy for Actinomycosis (HEYERDAHL (N), 1914, 1915, 1916, 1919, 1925, 1927, ENGELSTAD (N),1932, 1933). X-ray therapy, attempted already a decade previously (NORDENTOFT (D), 1914 b), became during the 1920-ties and 1930-ties the principal means for the treatment of the infection (EIKEN (D), 1924, and onwards, LINDGREN (F), 1931, RENANDER (S), 1937, J. NIELSEN (N), 1939), until, in 1939, the first report on a succesful treatment with Sulphonamide, M & B 693, appeared (MOENE (N)).

NOCARDIOSIS

Slides with scales containing *Microsporon minutissimum* (*Nocar-*

dia minutissima) were demonstrated, and the staining method discussed by C. Boeck (N) in 1885 and 1887 (b).

For the treatment of Erythrasma Rasch (D) (1905, 1909, 1927) had recommended alcoholic Iodine, or aqueous Potassium permanganate solution, as well as Sulphur, Resorcin, and Pyrogallic ointment. Lomholt (D), in 1939, however, followed the same lines of treatment as indicated for Pityriasis versicolor.

From 1926 (a) there exists a report by Reenstierna (S) on an experimental reproduction of Mycetoma in rabbits with strains of *Nocardia madurae*, isolated at the Pasteur Institute in Tunis.

ACID-FAST ELEMENTS AND FUNGI

In 1912/13 Reenstierna (S) forwarded a hypothesis on the mycotic nature of the microorganisms of Leprosy and Tuberculosis. He based this hypothesis on the occurrence of these microorganisms in polymorphic, streptococc-, and diphteroid-like non acid-fast forms. Later, in 1937, he stated that his non acid-fast strain, cultivated in 1912 from the blood of a patient suffering from classical nodular Leprosy, had shown an ability to change, partially, into acid-fast forms — in cultures as well as after inoculation into a monkey. In the culture there appeared also acid-fast egg-shaped formations.

In 1926 (b) Reenstierna (S) stated that he, since 1912, had also studied the behaviour of a yeast-like organism, morphologically resembling an *Oidium* (probably *Candida albicans*), and found it to yield acid-fast rods in one cell cultures. The author assumed that this yeast-like organism constitutes the mycotic, non acid-fast phase of the aetiological agents of Leprosy and Tuberculosis.

The study of the relations of the yeast-like fungus to the Bacillus of Koch was continued by Reenstierna's disciples, Gullberg (S), 1938, and Hallberg (S), 1940.

In 1917 Schaumann (S) came with new viewpoints on the aetiology of Lymphogranuloma benigna (LB). Taking in part into account Reenstierna's hypothesis on a possible relationship between the Leprosy as well as the Tuberculosis microorganisms and fungi, Schaumann (S), on the basis of analogous findings of pleomorphic *Streptothrix*, Diphteroid, and Coccus-like organisms in cultures from lymphatic glands in L.B., pointed out the possible mycotic nature of the disease. In addition, in 1934, he stated that serological complement-fixation reactions with antigens from the Diphteroid and *Streptothrix bacilli* yielded a manifest parallelism with Besredka's Tuberculosis antigen.

Since 1917 Schaumann (S) had also carried on with a research into certain peculiar corpuscules present in the tissue of L.B., which corpuscules he believed to be the product of a possible transformation of the Bacilllus of Koch into elements of mycotic nature.

In 1931—1932 Møllgaard (D) stated that he had succeeded, by cultivating KOCH's Bacillus, in obtaining Gram-positive non acid-fast coryneform Diplococci, as well as actinophytic threads with lateral sprouts, and fragmental conidia. These lateral sprouts, as well as the fragmentary conidia could display acid-fastness. The transformation was to a great deal reversible. MØLLGAARD (D) met with such Actinomyces-like forms in abundance also in calves and rabbits infected experimentally with cultures of KOCH's Bacillus.

SEROLOGY AND IMMUNOLOGY

HARBITZ & GRØNDAHL (N), in 1910, carried out experiments on agglutination and complement fixation reactions with Actinomycetes. But the results achieved were contradictory, and the reactions not applicable for diagnostical purposes.

In 1930 HOLM (D) found that 9 strains of man-pathogenic anaerobic Actinomycetes with regard to their agglutination proporties, fell into two distinct groups.

BØE, HARTMANN & THJØTTA (N), in 1939, published a serological study with rabbit sera of 10 *Aspergillus fumigatus* strains, isolated from the lungs of dead grouse. In complement fixation reactions titers up to a dilution of 1:1,600 were registered. Precpitation tests gave only low titers. The agglutination test, however, could not be used on account of spontaneous reactions in all plain rabbit sera. No difference was found between spores and mycelium as to antigenic effect. Neither could a serological differentiation be made between the strains.

LABORATORY AND DIAGNOSTICAL TECHNIQUES

C. BOECK (N), in 1885, demonstrated the use of EHRLICH-WEIGERT's Gentianaviolet solution for the detection of fungi in samples from skin lesions. But later, in 1887 (b), after having worked personally with WEIGERT, and on that occasion having tried out SAHLI's Methylenblue solution with Borax on Favus, he, henceforth recommended SAHLI's solution for the above-mentioned purpose. Whereas EHRLICH-WEIGERT's solution had stained even the surrounding tissues, SAHLI's stain made the coloured fungous elements appear on an almost uncoloured background. Moreover, in 1894, BOECK (N) advised to apply the solution directly to the skin, the stains making the scaling lesions stand out as strongly coloured patches against an only slightly tinged healthy skin.

ØRSKOV (D), in 1922, developed an agar-block method for the isolation of bacteria in pure culture from single cells, and a procedure for the direct tracing of bacterial growth. The method proved to ØRSKOV (D) of particular value in his investigations into the morphology of ray fungi (1923).

In 1928 MAGNUSSON (S)) introduced HUDDLESON's carbon dioxide gas method for isolation of Actinomycetes in LÖFFLER's serum tubes.

PALM & HINRICSSON (S), in 1940, recommended an impression-method for field diagnostics of parasitic incl. fungal skin disorders. Samples were collected by impressing a collodium film against the lesions. Then the film was examined microscopically, either in natural or coloured state.

ACKNOWLEDGEMENTS

The author wishes to express his thanks to Prof. R. VANBREUSEGHEM, General Secretary of ISHAM, and Prof. T. BENEDEK, Editor-in-Chief of this Journal, for proposing him to write the above paper. The author is grateful to Prof. SVEN HELLERSTRÖM, Head of the Dept. of Dermatology, Karolinska Institutet, Stockholm, for helping him to overcome the technical as well as the financial difficulties connected with the compiling of the manuscript. And last but not least the author is indebted to his sister Dr. Aime RINNE, and her husband Civ. Ing. O. RINNE, Sweden, who, by offering him economical support, made it possible for him to fulfil the task.

BIBLIOGRAPHY

Within () indicates the actual year of the announcement.

ÄYRÄPÄÄ, Matti (1893a) Actinomycosis. (Lecture with case report). Anniversary of Duodecim, 18.11.1893. Duodecim, **10**, 1, 6—15, 1894.

ÄYRÄPÄÄ, Matti (1893b) Actinomycosis. (Review with two case reports. Publ. in extenso in Skand. Tandläkarefören. Tidskr. **3**, 1, 1894). Finska Tandläkaresällskapets sammanträde, 18.12.1893. Skand. Tandläkarefören. Tidskr. **3**, 4, 235, 1894.

ÄYRÄPÄÄ, Matti (1894a) Actinomycosis. Skand. Tandläkarefören. Tidskr. **3**, 1, 1—12, 1894.

ÄYRÄPÄÄ, Matti (1894b) Aktinomykosis. (Lecture). Svenska Tandläkare-Sällsk. Förhandl. 1894. 25.1.1894. Skand. Tandläkarefören. Tidskr. **3**, 2, 94, 1894.

AFZELIUS, Arvid (1915) Handbok i Hudsjukdomarnas behandling. (Manual for the treatment of skin diseases). A. B. Nordiska Bokhandeln, Stockholm, 1915. Pp. 178.

AFZELIUS, Arvid (1925) Handbok i Hudsjukdomarnas behandling. Andra genomsedda upplagan. (Manual for the treatment of skin diseases. Second revised edition). Nordiska Bokhandelns Förlag, Stockholm, 1925. Pp. 225.

AHLSTRÖM, E. (1930) Ein Fall von Aktinomykose in den Adnexen der weiblichen Genitalien. Festskrift tillägnad Professor Elis ESSEN-MÖLLER. Acta obstet. gynec. Scand. **9**. 1—4, 1—29, 1930.

AHLTORP, Gideon (1933) Ett fall av aktinomykos i bäckorganen hos kvinna. (A case of Actinomycosis of the female pelvic organs). Hygiea, **95**, 2, 33—44, 1933.

ALMKVIST, J. (1924) Un cas de Achorion Quinckeanum (favus des rats) scroti. Soc. Derm. Stockholm pendant l'année 1924. Séance, 10.12.1924. Acta derm.-vener. **6**, 4, 555, 1925.

ALMKVIST, J. (1925) Eruption scrotale rappelant celles des moisissures. (Case. Same patient as Acta derm.-verer. **6**, 555, 1925). Soc. Derm. Stockholm pour l'année 1925. Séance, 14.1.1925. Acta derm.-vener. **8**, 1, 36, 1927—1928.

ALMQUIST, Ernst (1890) Untersuchungen über einige Bacteriengattungen mit Mycelien. Z. Hyg. **8**, 189—197, 1890.

ALMQUIST, E(rnst) (1893) Critical abstract on "Om Sop paa levande Jordbund" by JOHAN-OLSEN, O., 1893. Hygiea, **55**, 9, 315—317, 1893.

ALMQUIST, (Ernst) (1904a) Fall af Herpes tonsurans maculosus et squamosus. (Case of Herpes tonsurans maculosus et squamosus). Derm. Sällsk. i Stockholm förhandl. år 1904. Sammankomst, 29.9.1904. Hygiea, Följd 2, **7,** 3, 305, 1907.

Almquist, (Ernst) (1904b) Favus disseminatus corporis. (Case.) Derm. Sällsk. i Stockholm förhandl. år 1904. Sammankomst, 24,11.1904. Hygiea, Följd 2, **7,** 3, 313, 1907.

Ammentorp, L. (1888) 4 Tilfälde af actinomycosis hominis. (Four cases of Actinomycosis hominis). Nordiskt Med. Ark. **20,** 23, 1—19, 1888.

Ammentorp, L. (1893a) Tilfaelde af Underlivsaktinomykose. (Case of Actinomycosis of the hypogastrium). Medicinsk Selskab. Møde, 7.3.1893. Hospitalstidende, **36,** 4 Raekke, **1,** 18, 518—520, 1893.

Ammentorp, L. (1893b) Om Aktinomykose. Kliniske Iagttagelser paa Kommunehospitalet. (On Actinomycosis. Clinical observations from the Municipal Hospital). Bibliotek for Laeger, **85,** 7 Raekke, **4,** 6, 433—472, 1893.

Andersen, Mildrid (1939a) Acne conglobata? Actinomycosis? (Case). Verh. d. Dänischen Dermatol. Ges. Sitzung, 4.10.1939. Acta derm.-vener. **21,** 4, 601—602, 1940, and Dansk Dermatologisk Selskabs Forhandlinger Sept. 1939-Maj 1940. 306 Møde, 4.10.1939. Nordisk Med., **9,** 1941 : 1, 11, Hospitalstidende, 825, 1941.

Andersen, Mildred (1939b) Genfremstilling af Pat. med Acne vulgaris? Actinomycose. (Re-demonstration of the patient with Acne vulgaris? Actinomycosis?) Dansk Dermatologisk Selskabs Forhandlinger Sept. 1939-Maj 1940. 310 Møde, 6.12.1939. Nordisk Med. **9,** 1941 : 1, 11, Hospitalstidende, 829, 1941.

Arndtsen, Adam (1851) Om en ny Planteform i Favus. (On a new plant species in Favus). Norsk Magazin for Laegevidenskaben, 2 Raekke, **5,** 316—328, 1851.

Backer Grøndahl, Nils (1910, 1911) See Harbitz, F. and Backer Grøndahl, Nils, 1910b, 1911a, 1911b.

Backer Grøndahl, N(ils) (1918) Tilfaelde av bukaktinomykose behandlet med vaccine. (Case of abdominal Actinomycosis treated with vaccine). Förhandlinger i Kristiania Kirurgiske Forening 1918, pp. 151—152. Møte, 16.9.1918. Norsk Mag. Laegevidensk. **80,** 5 Raekke, **17,** Bilaga, 1919.

Bäärenhielm, G. (1900) Ett fall af appendicitis actinomycotica. (A case of Appendicitis actinomycotica). Hygiea, **62,** 5, 545—550, 1900.

Bang, B. (1883a) Aktinomykosen eller Straalesvampesygdommen. (Actinomycosis). Tidsskr. f. Veterinaerer, København, 2 Raekke, **13,** 47—110, 1883.

Bang, B. (1883b) Aktinomykosen eller Straalesvampesygdommen. (Actinomycosis). Hospitals-Tidende, 3 Raekke, **1,** 29, 673—680; 30, 697—706; 31, 721—730, 1883.

Bang, B. (1884) Die Strahlenpilzerkrankung (Actinomycosis). Dtsch. Z. Tiermed. **10,** 249—264, 1883—1884.

Bang, Hans Olaf (1940) Nogle kliniske B_1-Vitaminanalyser ved Hjaelp af Svampemetoden. (Some clinical B_1-Vitamine analyses with the fungus method). Ugeskrift for Laeger, **102,** 39, 1000—1002, 1940.

Bang, Henrik (1907) Notiser om en Mikrospori-Epidemi paa Amager. (Comments on a Microsporia epidemic at Amagar). Hospitalstidende, **50,** 4 Raekke, **15,** 47, 1254—1260, 1907.

Bang, Henrik (1910a) Sur une Trichophytie cutanée à grandes cercles, causée par un Dermatophyte nouveau (Trichophyton purpureum, Bang). Ann. Dermatol. Syphiligr., 5e Sér., **1,** 5, 225—238, 1910.

Bang, (Henrik) (1910b) Die dänischen Trichophyten und ihre Stellung in Sabouraud's System. Verh. 1. Kongr. Nord. Dermatol. Ver., Kopenhagen, 17—19 Mai 1910, pp. 9—18. Nordiskt Med. Ark., Afdelning 2, Anhang, 1911.

Bang, (Henrik) (1910c) Les trichophytons en Denmark. (Abstract by the author). Société scandinaire de Dermatologie, mai 1910. Ann. Dermatol. Syphiligr. 5e Sér., **1,** 8, 415, 1910.

Bang, Henrik (1910d) Nyere Undersøgelser over Hudens Svampesygdomme. (Newer investigations into the fungous diseases of the skin. Survey). Hospitalstidende, **53,** 5 Raekke, **3,** 28, 835—839, 29, 857—863, 1910.

Bang, H(enrik) (1910e) Samtidig Forekomst af Favus of Trichophyti i Haarbunden. (Simultaneous occurrence of Favus and Trichophytosis in the capillitium. Case). Dansk Dermatologisk Selskab. 77 Møde, 7.12.1910. Hospitalstidende, **54,** 5 Raekke, **4,** 24, 675, 1911.

Bang, (Henrik) (1913) Favus, trichophyti, mikrospori. (Manuskript). (Favus, Trichophytosis, Microsporisa. Dermatophytes met with in Denmark from 1909 to 1913. Manuscript). Quoted by Sylvest, B., Acta derm.-vener. **29**, 225, 1949.

Bang, H(enrik) & Haslund, Otto (1915) Et Tilfaelde af Trikophytia ungium et capillitii et corporis med Kultur af Trichophyton violaceum. (A case of Trichophytia ungium et capillitii et corporis yielding a culture of Trichophyton violaceum). Hospitalstidende, **58,** 5 Raekke, **8,** 4, 81—89, 1915.

Bang, Henrik (1921) Et Tilfaelde af Skaegtrikofyti behandlet med intravenøse Jod-Jodkalium injektioner (efter Ravaut). (A case of Trichophytosis in the beard treated with intravenous Potassium iodide injections, according to Ravaut) Dansk Derm. Selskabs Forhandl. 1921—1922, pp. 5—7. 159 Møde, 5.10.1921. Hospitalstidende, **65,** Bilaga, 1922.

Bang, Henrik (1923) Et Tilfaelde af Sekundaere Trichophytider med morbilliformet Udslet. (A case of secondary Trichophytids with a morbilliform dissemination). Dansk Derm. Selskabs Forhandl. 1923—1924, pp. 13—14. 177 Møde, 7.11.1923. Hospitalstidende, **67,** Bilaga, 1924.

Bang, Henrik (1937) Behandling af Mikrospori i Haarbunden med Mycocten. (Treatment of Microsporia in the capillitium with Mycocten). Jydsk Med. Selskabs Forhandl. 1936—1937, pp. 20—23. 127 Møde, 8.11.1937. Hospitalstidende, **80,** Bilaga, 1937.

Bengtson, H. (1923) Ringorm hos häst. (Ringworm in horse). Svensk vet. Tidskr. **28,** 8—9, 277—281, 1923.

Bentzen, G. E. (1890) Nogle bemerkninger om trøske, navlig dens idiopatiske optraeden hos voksne. (Some remarks on Thrush, viz. its idiopathical occurrence in adults). Tidsskr. Norske Laegeforening, **10,** 4, 162—167, 1890.

Berg, (Frederik Theodor) (1841a) Förevisning av Aphthae under mikroskopet, med discussion on denna sjukdoms natur. (Microscopical demonstration of Aphthae, with a discussion on the nature of this disease. Publ. as "Torsk i mikroskopiskt anatomiskt hänseende" i Hygiea, **3,** 12, December, 1841). Årsberättelse om Svenska Läkare-Sällskapets arbeten. Lemnad den 5 October 1841, p. 70.

Berg, (Frederik Theodor) (1841b) Meddelande. (Announcement of a communication on the mycotic nature of Aphthae, to be given on the 30.11.1841). Sammankomst, 23.11.1841. Förhandlingar vid Svenska Läkare-Sällskapets sammankomster från och med 5 Oct. 1841 till och med 26 Sept. 1843, p. 7.

Berg, (Frederik Theodor) (1841c) Föredrag och mikroskopisk visning av aphtae, samt mögelsvampar. (Lecture and microscopical demonstration of Aphthae and mould fungi). Sammankomst, 30.11.1841. Förhandlingar vid Svenska Läkare-Sällskapets sammankomster från och med 5 Oct. 1841 till och med 26 Sept. 1843, p 7.

Berg, F(rederik) T(heodor) (1841d) Torsk i mikroskopiskt anatomiskt hänseende. (Thrush from the microscopical anatomical point of view). Hygiea, **3,** 12, 541—550, 1841.

Berg, (Frederik Theodor) (1842a) Lettre de M.le Docteur Berg à M.le Docteur Gruby, sur la structure du muguet. Stockholm, le 20 mai 1842. La Clinique des Hôpitaux des Enfants, 2e année, 143, 1842.

Berg, F(rederik) T(heodor) (1842b) De la structure anatomo-microscopique du muguet. La Clinique des Hôpitaux des Enfants, 2e année, 143—145, 1842.

Berg, F(rederik) T(heodor) (1842c) Om naturen af Torsk, Aphthae. (On the nature of the Thrush). Förhandlingar vid De Skandinaviske Naturforskarens tredje Möte i Stockholm, 13—19 Juli 1842, p. 769.

Berg, Fr(ederik) Th(eodor) (1842d) Årsberättelse öfver späda barnens helso och sjukvård på Stockholms Allmänna Barnhus år 1842. (Annual report for the year 1842 on the state of health and the ward of infants at Stockholm's Public Orphanage). Svenska Läkare-Sällsk. nya Handl. **3,** 20—128, 1843.

Berg, Fr(ederik) Th(eodor) (1843) Årsberättelse öfver späda barnens helso-och sjukvård på Stockholms Allmänna Barnhus år 1843. (Annual report for the year 1843 on the state of health and the ward of infants at Stockholm's Public Orphanage). Svenska Läkare-Sällsk. nya Handl. **4,** 185—339, 1844.

Berg, (Frederik Theodor) (1844) Favus hos ett dibarn. (Favus in an infant. Case.) Svenska Läkare-Sällskapets sammankomst, 16.4.1844. Förhandlingar vid Svenska Läkare-Sällskapets sammankomster från och med Octob. 1843 till och med Sept. 1844, p. 54.

Berg, F(rederik) T(heodor) (1846a) Om Torsk, serdeles i barnaåldern. (On Thrush, particularly in childhood). Hygiea, **8,** 6, 337—352; 7, 401—422; 8, 465—488; 9, 529—544; 10, 593—608; 11, 657—674; 12, 721—760, 1846.

Berg. Fr(ederik) Th(eodor) (1846b) Om Torsk hos barn. (On Thrush in children). L. J. Hjerta, Stockholm, 1846, Pp. 157.

B(erg, J.) (1890) Actinomycosis. (Pp. 9—11). Rapport för år 1890 från Kongl. Serafimerlasarettets Kirurgiska Afdelning, av Rossander, Carl. J.

Berg, Sigurd (1930) Två fall av lungaktinomykos. (Two cases of pulmonary Actinomycosis). Tuberkuloseläkareföreningens Förhandlingar 1930. Sammanträde, 8.10.1930. Hygiea, **93,** 8, 319—325, 1931.

Bergenhem, Bengt, (1929) Bidrag till kännedomen om aktinomykosen, speciellt dess bakteriologiska diagnostik, med särskild hänsyn tagen till svenska arbeten under senare tid. (Contributions to the knowledge of Actinomycosis, especially its bacteriological diagnosis, with particular regard to Swedish publications of later date). Festskrift tillägnad Professor Ulrik Quensel Upsala Läk Fören. Förh., Ny följd, **34,** 1—2, 421—458, 1928—1929.

Bergh, R. (1863) Om constitutional Syphilis, paadraget uden Samleie, Syphilis insontium. (On constitutional Syphilis, aquired without cohabitation. Syphilis insontium. Footnote on Pityriasis versicolor, p. 50). Hospitals-Tidende, **6,** 13, 49—51, 1863.

Bergh, R. (1869a) Tilfaelde af Negleskurv, favøs Onychomycose. (Case of nail Favus, favic Onychomycosis). Hospitals-Tidende, **12,** 23, 89—90; 24, 93—94, 1869.

Bergh, R. (1869b) Beretning fra Almindelig Hospitals anden Afdelning for 1869. (Report for 1869 from Second Department of the General Hospital). Hospitals-Tidende, **13,** (Pp 153—154), 1870.

Bergh, R. (1870) (Same for 1870). Hospitals-Tidende, **14,** (P. 129), 1871.

Bergh, R. (1871) (Same for 1871). Hospitals-Tidende, **15,** (P. 149), 1872.

Bergh, R. (1872) (Same for 1872). Hospitals-Tidende, **16,** (Pp. 157—158), 1783.

Bergh, R. (1873) (Same for 1873). Hospitals-Tidende, 2 Raekke, **1,** 41, (Pp. 645—647), 1874.

Bergh, R. (1874) (Same for 1874). Hospitals-Tidende, 2 Raekke, **2,** 29, (Pp. 450—452), 1875.

Bergh, R. (1875) (Same for 1875). Hospitals-Tidende, 2, Raekke, **3,** 36, (Pp. 561—564), 1876.

Bergh, R. (1876) (Same for 1876). Hospitals-Tidende, 2 Raekke, **4,** 34, (Pp. 542—545), 1877.

Bergh, R. (1877) (Same for 1877). Hospitals-Tidende, 2 Raekke, **5,** 38, (Pp. 604—607), 1878.

Bergh, R. (1878) (Same for 1878). Hospitals-Tidende, 2 Raekke, **6,** 40, (Pp. 802—805), 1879.

Bergh, R. (1879) (Same for 1879). Hospitals-Tidende, 2, Raekke, **7,** 30, (Pp. 581—584), 1880.

Bergh, R. (1880) (Same for 1880). Hospitals-Tidende, 2 Raekke, **8,** 36, (Pp. 703—705), 1881.

Bergh, R. (1881) (Same for 1881). Hospitals-Tidende, 2 Raekke, **9,** 34, (Pp. 676—678), 1882.

Bergh, R. (1882) (Same for 1882). Hospitals-Tidende, 3 Raekke, **1,** 26, (Pp. 609—611), 1883.

Bergh, R. (1883) (Same for 1883). Hospitals-Tidende, 3 Raekke, **2,** 42, (P. 991), 1884.

Bergh, R. (1884) (Same for 1884). Hospitals-Tidende, 3 Raekke, **3,** 37, (P. 977), 1885.

Bergh, R. (1885) (Same for 1885). Hospitals-Tidende, 3 Raekke, **4,** 40, (P. 946), 1886.

Bergh, R. (1894) Mykosis versicolor hos Smaabørn. (Mycosis versicolor in infants). Hospitals-Tidende, **37,** 4 Raekke, **2,** 35, 863, 1894.

Bergholm, Hjalmar (1902) Ueber Mikroorganismen des Vaginalsecretes Schwangerer. Arch. Gynaek. **66,** 3, 497—589, 1902.

Bergman, Rolf (1929a) Mögelsvampinfektion i lungan. (Mould fungus infection of the lung. Demonstration of pathological preparations. Publ. in Tidskr. i Militär Hälsovård, **54,** 4, 209—229, 1929). Svenska Militärläkareföreningens förhandlingar. Stockholmsavdelningens sammanträde, 17.10.1929. Tidskr. i Militär Hälsovård, **54,** 3, 186—187, 1929.

Bergman, Rolf (1929b) Om mögelsvampinfektioner i lungan. Zusammenfassung. (On mould fungus infections of the lung. Same as Bergman, Rolf & Henschen, F. in Beitr. Klin. Tuberk. **73,** 467, 1929/30). Tidskr. i Militär Hälsovård, **54,** 4, 209—229, 1929.

Bergman, Rolf & Henschen, Folke (1930) Zur Kasuistik der Lungenaspergillose. Beitr. Klin. Tuber., **73,** 463—484, 1929/30.

Bergman, R(olf) (1933a) Diskussionsinlägg till Danielson, C. G., Fall av pneumonomykos. (Contribution to a discussion. Case of Pneumomycosis). Förhandlingar vid Svenska Läkaresällskapets sammankomster år 1933.

Bergman, Rolf, Danielson, C. G., Ottander, Olle & Schnell, Åke (1933b) Ytterligare bidrag till kännedomen om lungmykoser i Sverige. 1. Fall med Aspergillus fumigatus (Bergman, Danielson, Schnell). 2. Fall av Oidiomykos (Ottander). (Further contribution to the knowledge of lung Mycoses in Sweden. 1. Case with Aspergillus fumigatus. 2. Case of Oidiomycosis). Festskrift till Professor Dr. Germund Wirgin. Nordisk Hyg. Tidskr., **14,** 1—5, 1—20, 1933.

Bidenkap (1885) Diskussionsinlägg. (Contribution to a discussion, concerning C. Boeck's demonstration of skin parasite preparations). Forhandlinger i det Norske Medicinske Selskab i Kristiania 1885, p. 155. Møde, 3.6.1885. Norsk Magazin for Laegevidenskaben, 3 Raekke, **15,** Bilaga, 1885.

Bidenkap, J. H. (1911) Trikofytien i Nordre Gudbrandsdalen. (Trichophytosis in the Northern Gudbrandsdal). Tidsskr. Norske Laegeforening, **31,** 10, 425—431, 1911.

Bjerrum, Oluf & Hansen, Svend (1932) Undersøgelser over Forekomsten af Aktinomyceter i Mundhulen og deres betydning for den akute Form af Aktinomykose. (Investigations into the occurrence of Actinomycetes in the oral cavity and their significance for the acute form of Actinomycosis). Ugeskrift for Laeger, **94,**, 45, 1075—1078, 1932.

Blegvad, N. Rh. (1908) Sort Tunge (Lingua villosa nigra). (Black tongue). Bibliotek for Laeger, **100,** 8 Raekke, **9,** 187—242, 1908.

Boas (1930a) Udbredt Dermatomykose i Tilslytning til Feberbehandling for Dementia paralytica. (Disseminated Dermatomycosis in connection with fever therapy of Dementia paralytica. Case). Dansk Dermatologisk Selskabs Forhandlinger Sept. 1929-Maj 1930, p. 60. 226 Møde, 2.4.1930. Hospitalstidende, **73,** Bilaga, 1930.

Boas (1930b) Kutan Trichophyti. (Cutaneous Trichophytosis. Cases). Dansk Dermatologisk Selskabs Forhandlinger Sept. 1930-Maj 1931, p. 17. 230 Møde, 3.12.1930. Hospitalstidende, **74,** Bilaga, 1931.

Böcher, Carl (1887) Lingua nigra. Hospitalstidende, **3,** Raekke, **5,** 10, 221—226, 1887.

Boeck, C. (1885) Demonstration af Preparater af Hudparasiter. (Demonstration of skin parasite preparations). Forhandlinger i det Norske Medicinske Selskab i Kristiania 1885, pp. 147—155. Møde, 3.6.1885. Norsk Mag. Laegevidensk., **3,** Raekke, **15,** Bilaga, 1885.

Boeck, C. (1887a) Meddelelse. (Report on Herpes tonsurans in cattle and sheep, and its transmittance to man). Forhandlinger i det Norske Medicinske Selskab i Kristiania 1887, pp. 100—101. Møde, 18.5.1887. Norsk Mag. Laegevidensk. **48,** 4 Raekke, **2,** Bilaga, 1887.

Boeck, C. (1887b) Fremvisning af Preparater af Hudparasiter. (Demonstration of skin parasite preparations). Forhandlinger i det Norske Medicinske Selskab i Kristiania 1887, pp. 119—123. Møde, 1.6.1887. Norsk Mag. Laegevidensk. **48,** 4 Raekke, **2,** Bilaga, 1887.

Boeck, C. (1888) Eczema circiné. Forhandlinger i det Norske Medicinske Selskab i Kristiania 1888, pp. 39—43. Møde, 22.2.1888. Norsk Mag. Laegevidensk. **49,** 4 Raekke, **3,** Bilaga, 1888.

Boeck, C. (1894) Diagnostisk farvemetode for Hyfsop paa Huden. (Staining method for diagnosis of Hyphomycetes on the skin). Forhandlinger i det Medicinske

Selskab i Kristiania 1894, pp. 57—59. Møde, 28.2.1894. Norsk Mag. Laegevidensk. **55,** 4 Raekke, **2,** Bilaga, 1894.

Boeck, C. (1914) Sporotrichosis. (Cases). Forhandlinger i det Medicinske Selskab i Kristiania 1914, pp. 150—152. Møde, 18.11.1914. Norsk Mag. Laegevidensk. **75,** 5 Raekke, **12,** Bilaga, 1914.

Boeck, W. (1852a) Klinik over Hudsygdommene og de syphilitiske Sygdomme. (Clinic on skin diseases and syphilitical disorders. Lecture. Favus, pp. 106—117). Norsk Mag. Laegevidensk. 2 Raekke, **6,** 94—124, 1852.

Boeck, W. (1852b) Klinik over Hudsygdommene og de syphilitiske Sygdomme i 1852. (Clinic on skin diseases and syphilitical disorders in 1852. Lecture. Favus, Pityriasis versicolor, pp. 217—230). Norsk Mag. Laegevidensk. 2 Raekke, **7,** 193—236, 1853.

Bøe, Johs., Hartmann, Otto & Thjøtta, Th. (1939) A Serological Study of Aspergillus fumigatus. Acta path. microbiol. scand., **16,** 2, 178—186, 1939.

Bonnevie, P(oul) (1933) 3 Tilfaelde af Onychomycose framkaldt af Scopulariopsis brevicaulis. (Three cases of Onychomycosis caused by Scopulariopsis brevicaulis). Dansk Derm. Selsk. Forhandl. Sept. 1933—Maj 1934, pp. 2—3. 4.10.1933. Hospitalstidende, **77,** Bilaga, 1934.

Bonnevie, Poul (1935) Experimental Investigations on the antimycotic Effect of Ethylparaoxybenzoate and of allied Substances. Proc. IX Meet. North. Derm. Soc., Copenhagen, 10—12 June. 1935. Acta derm.-vener., **17,** 5, 576—588, 1936.

Bonnevie, Poul (1939a) Aetiologie und Pathogenese der Ekzemkrankheiten. Klinische Studien über die Ursachen der Ekzeme unter besonderer Berücksichtigung des diagnostischen Wertes der Ekzemproben. 3. Das mykotische Ekzem. (Pp. 451—483). Thesis, Copenhagen, 1939. Nyt Nordisk Forlag, Arnold Busck, Kopenhagen, und Johann Ambrosius Barth, Verlagsbuchhandlung, Leipzig, 1939. Pp. 600.

Bonnevie, P(oul) (1939b) Overføldsomhed for Aetylparaoxybenzoat (Mycocten). (Sensibility for Ethylparaoxybenzoate. Mycocten). Dansk Dermatologisk Selskabs Forhandlinger. 303 Møde, 1.3.1939. Nordisk Med. **6,** 1940: 2, 14, Hospitalstidende, 684—685, 1940.

Bonsdorff, von Hj. (1892) Förevisning av tvenne pat. lidande av aktinomykos. (Demonstration of two patients suffering from Actinomycosis). Finska Läkaresällskapets möte, 24.9.1892. Finska Läkaresällsk. Handl. **34,** 10, 799, 1892.

Bonsdorff, von, Hj. (1894) Actinomycosis hominis. Ett bidrag till kännedomen om denna sjukdoms förekomst i Finland. (Actinomycosis hominis. A contribution to the knowledge of the occurrence of this disease in Finland). Finska Läkaresällsk. Handl. **36,** 4, 253—298; 6, 395—446, 1894.

Bonsdorff, von Hj. (1901) Torakal aktinomykos. (Thoracic Actinomycosis. Four cases). Finska Läkaresällskapets sammanträde, 23.2.1901. Finska Läkaresällsk. Handl. **43,** 5, 393, 1901.

Borch, G. F. (1865) Bidrag till Laeren om de planteparasitiske Hudsygdomme. (Contribution to the principles of plant-parasitic skin diseases). Thesis, Copenhagen, 1865. J. R. Veldby, Kjøbenhavn, 1865. Pp. 110.

Borelius, Jacques (1896a) Om aktinomykos i Blekinge. (On Actinomycosis in Blekinge). Sydöstra Sveriges läkare och apotekare föreningens möte, 12. 4.1896. Hygiea, **58,** 8, 156—164, 1896.

Borelius, Jacques (1896b) Genmäle till dr. G. Ekehorn. (Reply to Dr. G. Ekehorn). Hygiea, **58,** 11, 548—549, 1896.

Borelius, Jacques (1903) Bauchaktinomykose in Form eines circumskripten beweglichen Tumors. Nordiskt Med. Ark. **36,** Afdelning 1, Kirurgi, 6, 1—16, 1903.

Brandt, Thore (1932) Report on an Epidemic of Trichophytosis. Proceedings of the Eights Meeting of the Northern Dermatological Society, Stockholm, 2—4 June 1932. Acta derm-vener., **13,** 5, 443—448, 1932.

Brofeldt, Simo (1921a) Om actinomykosen i Finland. (On Actinomycosis in Finland). Forhandlinger ved Nordisk Kirurgisk Forenings 13 : e Møte i Helsingfors, 7—9 Juli 1921, pp. 40—41.

Brofeldt, (Simo) (1921b) Sur l'actinomycose en Finlande. 13e Congres Scandinave de Chirurgie à Helsingfors (Finlande), 7—9 juillet 1921. Acta chir. Scand., **55,** 167, 1922/23.

Brofeldt, (Simo) (1926) Über Aktinomykose in Finland. Acta Soc. Med. Fenn. "Duodecim" **7**, 6, 1—41, 1926.

Brotherus, W. E. (1913) Eräs streptotrikositapaus sekä yleinen katsaus streptotrikseihin. Deutsches Referat: Ein fall von Streptotrikose samt einer Übersicht über die Streptotrikosen. Duodecim, **29**, 6, 299—340, 1913.

Brun Pedersen, M. (1916a) Om de saakaldte sekundaere likenoide Trikofytider (On so-called secondary lichenoid Trichophytids). Hospitalstidende, **59**, 5 Raekke **9**, 20, 465—481, 1916.

Brun Pedersen, (M). (1916b) Sekundaere likenoide Trikofytider. (Secondary lichenoid Trichophytids). Dansk Dermatologisk Selskab. 113 Møde, 2.2.1916. Hospitalstidende, **59**, 5 Raekke, **9**, 33, 819—820,1916.

Brun Pedersen, M. (1916c) Udbredt Eczema marginatum efter Omslagsbehandling. (Disseminated Eczema marginatum in consequence of treatment with wet dressings). Ugeskrift for Laeger, **78**, 27, 1131—1133, 1916.

Brun pedersen, M. (1917) Ein Beitrag zum Studium der Pathogenese der sekundären Trichophytide. Derm. Z. **24**, 12, 731—736, 1917.

Brun Pedersen, (M), (1918) Sekundaere likenoide Trikofytider. (Secondary lichenoid Trichophytids). Dansk Dermatologisk Selskab. 127 Møde, 6.2.1918. Hospitalstidende, **61**, 40, 1366—1367, 1918.

Bruun, Egon (1937) Et Tilfaelde af Aktinomykose-pyaemi. (A case of Actinomyces-pyaemia). Kirurgisk Selskab, 16.10.1937. Hospitalstidende, **81**, 1, 16—20, 1938.

Bruusgaard, E. (1912) Sporotrikose. En oversigt. (Sporotrichosis. A survey). Norsk Mag. Laegevidensk. **73**, 5 Raekke, **10**, 1085—1900, 1912.

Bruusgaard, E. (1919) Kasuistiske meddelelser fra Rikshospitalets avdelning for hudsygdomme. 2. Lichenoid trikofyti. (Casuistical communications from the department of skin diseases at the Riks-Hospital. 2. Lichenoid Trichophytosis, pp. 136—137). Tidsskr. Norske Laegeforening, **39**, 4, 134—137, 1919.

Bruusgaard, E. (1920) Dyp trikofyti i skjegget med haematogene utbrud paa huden. (Deep Trichophytosis of the beard, with a haematogenous rash on the skin). Fordhandlinger i det Medicinske Selskab i Kristiania 1920, pp. 49—50. Møte, 10.3.1920. Norsk Mag. Laegevidensk. **81**, 5 Raekke, **18**, Bilaga, 1920.

Bruusgaard, E. (1921) Über hämatogene Trichophytie. Verh. dtsch. Derm. Ges. Zwölfter Kongr. Hamburg, 17—21 Mai 1921. Arch. Derm. Syph. **138**, 381—387, 1922.

Bruusgaard, E. (1922a) Om haematogen trikofyti. (On haematogenous Trichophytosis). Norsk Mag. Laegevidensk. **83**, 5 Raekke, **20**, 2, 83—88, 1922.

Bruusgaard, E. (1922b) Haematogenous Infection in Trichophytia. Acta Derm.-vener. **3**, 1—2, 21—31, 1922.

Cappelen, A. (1896a) 2 Patienter med Aktinomykose i Overkjaeven og Kindet. (Two patients with Actinomycosis of the upper jaw and the cheek. Cases). Forhandl. Med. Selsk. i Kristiania 1896, pp. 85—86. Møde, 11.3.1896. Norsk Mag. Laegevidensk. **57**, 4 Raekke, **11**, Bilaga, 1896.

Cappelen, (A.) (1896b) Actinomykose. (Actinomycosis. Re-demonstration of a case). Forhandlinger i det Medicinske Selskab i Kristiania 1896, p. 125. Møde, 20.5.1896. Norsk Mag. Laegevidensk. **57**, 4 Raekke, **11**, Bilaga, 1896.

Christensen, A. & Eiken, Th. (1924) Om Röntgenbehandling af Aktinomykose. (On X-ray treatment of Actinomycosis). Ugeskrift for Laeger, **86**, 49, 937—939, 1924.

Christiansen, M. (1922) General Mucormycose hos Svin. (General Mucormycosis in pig). Kgl. Veterinaer- og Landbojhøjskols Aarsskr. **6**, 133—190, 1922.

Christiansen, M. & Nielsen, Nils (1929) Mucormycose beim Schwein. Virchow's Arch. **273**, 3, 829—863, 1929.

Conradi (1887) Tumer pulmonum, Aktinomykose eller Sarkom, kronisk Pyaemi. Sektionsfund og mikroskopisk undersøkelse av Axel Holst. (Tumor pulmonum, Actinomycosis or Sarcoma, chronic Pyaemia. Post-mortem finds and microscopical examination by Axel Holst). Forhandlinger i det Norske Medicinske Selskab i Kristiania 1887, pp. 101—106. Møde, 18.5.1887. Norsk Mag. Laegevidensk. **48**, 4 Raekke, **2**, Bilaga, 1887.

Cygnaeus, W. Quoted by Äyräpää, M. (P.7) in Skand. Tandläkareföreningens

Tidskr. **3,** 1, 1894, and by Bonsdorff, v., H. (P.298) in Finska Läkaresällsk. Handl. **36,** 253, 1894.

Dahl, Johan (1888) Om Favus i Buskeruds Amt. (On Favus in the District of Buskerud). Norsk Mag. Laegevidensk. **49,** 4 Raekke, **3,** 392—395, 1888.

Dalén, Albin (1902) Smärre oftalmologiska meddelanden. b) Tva fall af Konkrement (aktinomyces) i canaliculus lacrymalis superior. (Shorter ophthalmological notices. b) Two cases of concretions, Actinomyces, in the canaliculus lacrimalis superior, pp. 176—184). Hygiea, Följd 2, **2,** 2, 172—184, 1902.

Danbolt, Niels (1938a) Tiefe kutane Moniliasis, ein tödlich verlaufender Fall. (Publ. in Acta derm.-vener. **21,** 98, 1940). Verh. Norw. Derm. Ges. 1938. Sitzung, 8.12.1938. Acta derm.-vener. **23,** 3, 302, 1941.

Danbolt, Niels (1938b) Deep cutaneous Moniliasis, a fatal case of a peculiar Type. Demonstrated before the Norwegian Derm. Soc. 2nd meeting, fall, 1938. Acta derm.-vener. **21,** 1, 98—117, 1940.

Danbolt, Niels and Mossige, Kjell (1938c) Silver foxes as source of infection with Trichophytia in man. Acta derm.—vener. **19,** 3, 368—374, 1938.

Danbolt, Niels and Mossige, Kjell (1939a) Sølvrev som smittekilde for Trichophyti hos mennesker. Summary: Silver Foxes as Source of Infectuen with Trichophytia. (Same as in Acta derm.-vener. **19,** 368, 1938). Norsk Vet. Tidsskr. **51,** 3, 85—92, 1939.

Danbolt, N(iels) (1939b) Silver Foxes as Source of Infection with Trichophytia in Man. (Abstract). Vet. J. **95,** 8, 326,1939.

Danbolt, N(iels) (1940) Actinomycosis. (Case). Meet. Norw. Derm. Soc. 12.9.1940. Acta derm.-vener., **24,** 4, 338, 1943/44.

Danielson, Carl Gustaf (1933a) Fall av pneumonomykos. (Case of Pneumomycosis). Sammankomst, 16.5.1933. Förhandl. vid Svenska Läkaresällskapets sammankomster år 1933, pp. 389—414.

Danielson, C(arl) G(ustaf) 1933b) See Bergman, R., Danielson, C.G., Ottander, O. & Schnell, Å., 1933b.

Danielssen, D.C. (1982) Planteparasitaere Hudsygdomme. (Plant-parasitic skin diseases). Samling af Iakttagelser over Hudens Sygdomme - Receuil d'Observations sur les Maladies de la Peau - ved W. Boeck og D.C. Danielssen, 4—5 Hefte. C. Floors Boghandel, Bergen, 1892. Pp. 80.

Dernby, K.G. (1923) See Naeslund, C. & Dernby, K.G., 1923.

Düben, von, Gustaf (1885) Ofversigt af de bidrag mikroskopet lemnat till den medicinska diagnostiken. I. Huden. l. Epiphyter. (A survey on the contributions the microscope has furnished to medical diagnostics. I. Skin. l. Epiphytes, pp. 8—16). Svenska Läkare-Sällsk. Nya Handl. **9,** 1—98, 1855.

Edberg, E. (1916) Ett fall av primär parotisaktinomykos hos ett 3-års barn (A case of primary Actinomycosis of the Parotis in a three-year-old child) Allmänna Svenska Läkaretindningen, **13,** 20, 571—575, 1916.

Ehlers, M. (1892) Trichophytie de la plante du pied. Société Francaise de Dermatologie et de Syphiligraphie. Séance, 8.12.1892. Ann. Dermatol. Syphiligr. 3 Sér., **3,** 1282, 1892.

Eiken, Th. (1924) See Christensen, A. & Eiken, Th., 1924.

Eiken, Th. (1926) Über die Röntgenbehandlung der Aktinomykose. Festskrift tillägnad Gösta FORSELL: 1. Acta Radiol., **6,** 391—398, 1926.

Eiken, Th. (1927) Om Diagnosen af Aktinomykose. (On the diagnosis of Actinomycosis). Jydsk Medicinsk Selskabs Forhandlinger 1927—1928, pp. 33—41. 74 Møde, 13.11.1927. Hospitalstidende, **71,** Bilaga, 1928.

Eiken, Th. (1928) Om Aktinomykose, med saerligt Henblik paa Diagnose og Behandling. (On Actinomycosis, with special regard to diagnosis and treatment). Dansk Kirurgisk Selskabs Forhandlinger 1928—1929, pp. 12—17. 130 Møde, 8.12.1928. Hospitalstidende, **72,** Bilaga, 1929.

Eiken, Th. (1929) Om Aktinomykose med saerligt Henblik paa Diagnose og Behandling. (On Actinomycosis, with special regard to diagnosis and treatment). Ugeskrift for Laeger, **91,** 28, 581—593, 1929.

Eiken, Th. (1930) Er kornholdigt Pus pathognomisk for Actinomykose? (Is granule-containing pus pathognomic for Actinomycosis?). Ugeskrift for Laeger, **92,** 25, 602—603, 1930.

Eiken, Th. (1939) Diskussion. (Contribution to a discussion concerning:) Nielsen, J., Om Røntgenbehandling af cervice-facial Aktinomykose. Dansk Radiol. Selsk. 165 Møde, 18.4.1939. Nordisk Med. ,**4,** 1939: 4, 52, Hospitalstidende, 3842, 1939.

Ekehorn, Gustaf (1895) Redogörelse för några fall af appendicit. Fall 1. Appendicitis actinomycotica. (Report on some cases of Appendicitis. Case 1. Appendicitis actinomycotica. Pp. 433—444). Hygiea, **57,** 5, 433—465, 1895.

Ekehorn, G(ustaf) (1896a) Berättelse för åren 1893—1896 från Härnösands lasarett. (Report for the years 1893—1896 from the Härnösand Hospital).

Ekehorn, Gustaf (1896b) Om aktinomykos i Blekinge. (On Actinomycosis in Blekinge. Concerning the paper by Borelius). Hygiea, **58,** 10, 422—424, 1896.

Ekehorn, G(ustaf) (1900) Ett fall af appendicitis (och typhlitis) actinomycotica; exstripation af appendix jämte resektion af coecum; helsa. (A case of Appendicitis, and Typhlitis, actinomycotica; extripation of the appendix together with the resection of the coecum; health). Upsala Läkarefören. Förh., Ny följd, **5,** 7, 455—459, 1899—1900.

Eklund, Fr. (1883) Contribution á l'étude du "Lepocolla repens" le champignon élémentaire du Psoriasis. Ann. Dermatol. Syphiligr. 2e Sér., **4,** 4, 197—203, 1883.

Eliasson, A. (1895) Om aktinomykos hos menniskan. Kritiska studier och erfarenheter från lasarettet i Ystad. Bilaga till årsrapporten från Lasarettet i Ystad för år 1895. (On Actinomycosis in man. Critical studies and experiences from the Ystad Hospital. Supplement to the annual report from the Ystad Hospital for 1895). Hygiea, **58,** 10, 337—358, 1896.

Eliasson, A. (1896) Två fall af aktinomykos, framgångsrikt behandlade med jodkali. Ur ämbetsberättelse för 1896 till medicinalstyrelsen. (Two cases of Actinomycosis treated successfully with Potassium iodide. From the official report for the year 1896 to the Board of Health). Eira **21,** 8; 262—264, 1897.

Emanuel, Sv. (1932) Trichophytia. (Cases). Dansk Dermatologisk Selskabs Forhandlinger Sept. 1932-Maj 1933, p. 6. 245 Møde, 2.11.1932. Hospitalstidende, **76,** Bilaga, 1933.

Engelstad (1855, 1859) See Hempel & Engelstad, 1855, 1859.

Engelstad, Rolf Bull (1932) Radiumbehandling av ansikts- og halsaktinomykose. (Radium treatment of Actinomycosis of face and neck). Norsk Mag. Laegevidensk. **93,** 2, 161—175, 1932.

Engelstad, Rolf Bull (1933a) Radiumbehandling av abdominal aktinomykose. (Radium treatment of abdominal Actinomycosis). Norsk Mag. Laegevidensk. **94,** 7, 759—763, 1933.

Engelstad, Rolf Bull (1933b) Radiumbehandlung von abdominaler Aktinomykose. Strahlenther. **47,** 547—550, 1933.

Engelsted, S. (1864) Kliniske Meddelelser fra Almindeligt Hospital. (Clinical communications from the General Hospital. Tinea favosa, Favus, pp. 57—58, Herpes circinatus, pp. 59—62). Otto Schwartz's Boghandel, Kjøbenhavn, 1864. Pp.191.

Engelsted, S. (1879) Klinisk Veiledning til Diagnose og Behandling af Hudens Sygdomme. De af Planteparasiter betingede Hudsygdomme. (Clinical guidance to the diagnosis and treatment of the diseases of the skin. Skin diseases caused by plant-parasites. Pp. 10—33). C. A. Reitzels Forlag, Kjøbenhavn, 1879. Pp. 394.

Engström, K. (1916) Herpes tonsurans hos djur och människor. (Herpes tonsurans in animal and man). Svensk Vet. Tidskr. **21,** 183—185, 1916.

Envald, Reinhold (1890) Vaikeasti parantuva Herpes tonsurans kummaksin käsiseljässä. (Obstinate Herpes tonsurans on the backs of both hands). Duodecim, **6,** 12, 204—206, 1890.

Envald, Reinhold (1891) Kynsien taudeista. (On nail affections). Duodecim, **7,** 4, 53—62, 1891.

Eriksson, Zaida (1934) Om bronkialastman med särskilt beaktande av den allergiska formen. Deutsches Referat: Über Asthma bronchiale mit besonderer Berücksichtigung der allergischen Form desselben. (Survey. Moulds, p. 47). Finska Läkaresällsk. Handl. **76,** 1, 29—56, 1934.

Essen, v. (1883) Aspergillus flavescens i hörselgången. (Report, read by af Schultén). Möte, 19.5.1883. Finska Läkaresällsk. Handl., **25,** 6, 398, 1883.

Fenger, C. E. (1868) Om det Virksomme ved Gjaerningen, Forraadnelsen og visse Arter af Sygdomssmitte. (On the active principle in fermentation, putrification, and certain kinds of infection). Hyg. Meddel. **5,** 127—203, 1866—68.

Fibiger, Johannes (1915) Adolph Hannover som Patolog. (Adolph Hannover as pathologist). Hospitalstidende, **58,** 5 Raekke, **8,** 449—470, 1915.

Finne, G. (1886) Om Smaasoppene. (On microfungi). Tidsskr. Prakt. Med. **6,** 9, 145—153, 1886.

Finsen, Hannes Chr. (S) (1924) Gaermycoser. (Yeast mycoses). Festskrift i Anledning af Dansk Dermatologisk Selskabs 25-Aars Bestaaen, 1898—1923. Bibliotek for Laeger, **116,** 5, 303—320, 1924.

Finsen, H(annes) C(hr).) S. (1925) Trichophytia barbae et colli. (Case). Dansk Derm. Selsk. Forhandl. 1924—1925, p. 55. 188 Møde, 1.4.1925. Hospitalstidende, **68,** Bilaga, 1925.

Finsen, H(annes) C(hr.) S. (1930) Epidermomykosis bei Säuglingen. C.R. séances. 8e Congr. Int. Derm. Syphiligr., Copenhague 5—9 août 1930, p. 585.

Fischer, Joh. Fred. (1907) Röntgenbehandling af Trikofyti. (X-ray treatment of Trichophytosis). Hospitalstidende, **50,** 4 Raekke, **15,** 48, 1297—1299, 1907.

Fischer, Joh. Fred. (1908) Röntgenbehandling af Trikofyti. (X-ray treatment of Trichophytosis). Hospitalstidende, **51,** 5 Raekke, **1,** 3, 68—71, 1908.

Flensburg, Carl (1916) Frederik Theodor Berg och prioriteten till torsk-svampens upptäckt. (Frederik Theodor Berg and the priority of the discovery of the Thrush fungus). Festskrift till Professor Dr. Erik Müller. Svenska Läkaresällsk. Handl. **42,** 3, 737—745, 1916.

Follum, L. (1888) Behandling af favus med borsyre. (Treatment of Favus with Boric acid). Tidsskr. Prakt. Med. **8,** 23, 550—551, 1888.

Fontenay de, O. E. (1875) Uddrag af Aarsberetningen fra Kjøbenhavns Amts-Sygehus for 1875. (Excerpt from the annual report for 1875 from the Municipal Hospital, Copenhagen). Hospitals-Tidende, 2 Raekke, **3,** 12, (Pp. 189—190), 1876.

Fontenay, de, O. E. (1876) (Same for 1876), Hospitals-Tidende, 2 Raekke, **4,** 17, (P. 268), 1877.

Fontenay, de, O. E. (1877) (Same for 1877), Hospitals-Tidende, 2 Raekke, **5,** 14, (P. 215), 1878.

Fontenay, de O. E. (1878) (Same vor 1878), Hospitals-Tidende, 2 Raekke, **6,** 9. (P. 171), 1879.

Fontenay, de O. E. (1879) (Same for 1879), Hospitals-Tidende, 2 Raekke, **7,** 18, (P. 347), 1880.

Fontenay, de O.E. (1880) (Same for 1880), Hospitals-Tidende, 2 Raekke, **8,** 18, (P. 349), 1881.

Fontenay, de O. E. (1881) (Same for 1881), Hospitals-Tidende, 2 Raekke, **9,** 29, (Pp. 594—597), 1882.

Forselles, af (1900) Actinomykos i munbotten. (Actinomycosis of the bottom of the oral cavity. Case). Finska Läkaresällsk. årsmöte 27.1.1900. Finska Läkaresällsk. Handl. **42,** 4, 499, 1900.

Friele, J. and Hartwig, Chr. (1894) Et tilfaelde af aktinomycosis. (A case of Actinomycosis). Medicinsk Rev. **11,** 355—358, 1894.

Fries, Oscar Robert (1867) Om svampbildningar på menniskans hud och deraf förorsakade sjukdomar. (Fungous formations on the skin of man, and the diseases caused by them). Edquist & Berglund, Upsala, 1867. Pp 40.

Friis, Hjalmar (1937) Ringormepizooti blandt Gardehusarregimentets Heste. (Ringworm epizootic among the horses of the Hussar Guard Regiment). Maanedsskr. f. Dyrlaeger, **48,** 305—309, 1936/37.

Friis, Ivar (1898) Lidt om Aktinomykose. (Something on Actinomycosis). Hospitalstidende, **41,** 4 Raekke, **6,** 44, 1153—1158, 1898.

Frölich, E. (1910) Et Tilfaelde af Underlivsaktinomykose. (A case of Actinomycosis of the hypogastrium). Hospitalstidende, **53,** 5 Raekke, **3,** 22, 601—614, 1910.

Gade, F. G. (1884) Actinomycosis bovis et hominis. (Survey). Norsk Mag. Laegevidensk. 3 Raekke, **14,** 5, 319—324, 1884.

Gade, F. G. (1886) See Johan-Olsen, O. & Gade, F. G., 1886b.

Geirsvold (& Harbitz) (1904) Blastomyceter og blastomycoser. (Blastomycetes and Blastomycoses. Demonstration. Pp. 299—301). Forhandl. Med. Selsk. i Kristiania 1904, pp. 295—301. Møde, 26.10.1904. Norsk Mag. Laegevidensk. **65,** 5 Raekke, **2,** Bilaga, 1904.

Gelbjerg-Hansen, G. (1930) Tilfaelde af Aktinomykose (Case of Actinomycosis). Dansk Derm. Selsk. Forhandl. Sept. 1930-Maj 1931, pp. 4—5.228 Møde, 1.10.1930. Hospitalstidende, **74,** Bilaga, 1931.

Genner, V. (1929) Et Tilfaelde af Trichofyti paa den ubehaarede Hud bestaaet i ca. 2 Aar samt 2 Tilfaelde af Negletrichofyti hos Patientens Moder og Søster. (A case of Trichophytosis on the glabrous skin of about two years' duration, and two cases of Trichophytosis of the nails in the patients mother and sister). Dansk Derm. Selsk. Forhandl. Sept. 1929-Maj 1930, pp. 20—21. 221 Møde, 6.11.1929. Hospitalstidende, **73,** Bilaga, 1930..

Genner, *i*V. (1932) Et haardnakket Tilfaelde af Trichophytia curtis. (A persistent case of Trichophytia cutis. Same as 6.11.1929). Dansk Derm. Selsk. Forhandl. Sept. 1931-Maj 1933, p. 43. 241 Møde, 2.3.1932. Hospitalstidende, **75,** Bilaga, 1932.

Gjessing, H. Chr. & Mossige, Kjell (1937) Epidermophytosis. Report of Cases in Three Brothers, One of Whom Showed a Hitherto Undescribed Clinical Type In the Scalp. Arch. Derm. Syphilol. **36,** 6, December, 1154—1157, 1937.

Grøn, Fr. (1915) Aktinomykose behandlet med JK. (Actinomycosis treated with Potassium iodide). Forhandl. Med. Selsk. i Kristiania 1915, pp. 222—224. Møte, 22.12.1915. Norsk Mag. Laegevidensk. **76,** 5 Raekke, **13,** Bilaga, 1915.

Grøn, Kristian (1894) Trøske i forløbet af forskjellige akute sygdomme hos voksne. (Thrush in the course of different acute diseases in adults). Tidsskr. Norske Laegeforening, **14,** 5, 99—103, 1894.

Grøn, Kr(istian) (1909) Sporotrichosis. Oversigt. (Survey). Medicinsk Rev. **26,** 4, 182—190, 1909.

Grøndahl (1910) Abdominalaktinomykose — behandled med Kulturer af Act. hominis. (Abdominal Actinomycosis — treated with cultures of Act. hominis). Forhandl. Med. Selsk. i Kristiania 1910, p. 214. Møde, 7.12.1910. Norsk Mag. Laegevidensk. **71,** 5 Raekke, **8,** Bilaga, 1910.

Gruby (1842) Réponse de M.le docteur Gruby à M. le Docteur Berg, médecin en chef de l'hôpital des Enfants à Stockholm. Paris, le 10 septembre 1842. La Clinique des Hopitaux des Enfants, 2e année, 191—192, 1842.

Gruby (1844) Recherches sur les cryptogames qui constituent la maladie contagieuse du cuir chevelu décrite sous le nom de Teigne tondante (Mahon), Herpes tonsurans (Cazenave). C. R. Acad. Sci., Paris, **18, 14,** 583—585, séance du 1 avril, 1844.

Grythe, Olaf (1933) Spiller visse sopparter en rolle for tannstensdannelse? (Do certain kinds of fungi play a role in the formation of tartar?) Norske Tannlaegeforen. Tidende, **43,** 1, 1—6, 1933.

Grythe, Olaf (1938a) Leptotrix. Experimentelle Untersuchungen an Leptotrix die aus klinisch gesunden Zahnfleisch gezüchtet wurden. Fabritius & Sønner, Oslo, 1938. Pp. 88.

Grythe, Olaf, (1938b) The role of Actinomyces and Leptotrix as an Etiological Factor in the Formation of Tartar and Salivary Calculi. Festskrift for Professor Johan Rygge. Norske Tannlaegeforen. Tidende, **48,** 433—447, 1938.

Grythe, Olaf, (1939a) Actinomycetene — deres forekomst i jorden og i varmblodige organismers legemer. (The Actinomycetes and their occurrence in soil and in the organisms of warm-blooded animals). Norske Tannlaegeforen. Tidende, **49,** 1, 16—25, 1939.

Grythe, Olaf (1939b) The Actinomycetes and their Occurrence in Soil and in the Organisms of Warm-Blooded Animals. Acta odont. Scand. **1,** 2, 155—170, 1939.

Grythe, Olaf (1941) Die Aktinomyzeten, ihr Vorkommen im Boden und im Körper der Warmblüter. Dtsch. Zahn-, Mund- und Kieferkunde, **8,** 1, 29—40, 1941.

Guldberg, Gustav (1932) Trichophytiene, deres ytringsformer og patogenese. (Trichophytoses, their manifestations and pathogenesis. Trial lecture). Tidsskr. Norske Laegeforening, **52,** 17, 1029—1047, 1932.

GULLBERG, Evert (1938) Some Observations Indicating the Possibility of a Relation of the Bacillus of KOCH to a Yeast-Like Fungus (of Oidium Type). A Posthumous Paper, Edited by J. REENSTIERNA in collaboration with R. HUSS, V. HALLBERG, G. HELLEBERG and C. G. LECZINSKY. Acta med. Scand. **94,** 4—6, 527—566, 1938.

GUNDERSEN, Edv. (1924, 1925a) See THJØTTA, Th & GUNDERSEN, Edv. 1924b, 1925.

GUNDERSEN, Edv. (1925b) Pilzuntersuchungen über das Problem der Gramfärbung. Verh. Dtsch. Derm. Ges. Vierzehnter Kongress gehalten zu Dresden, 13—16 September 1925. Arch. Derm. Syphilis, **151,** Kongressbericht, 171—175, 1926.

GUNDERSEN, Edv. (1926a) Pilzuntersuchungen über das Problem der Gramfärbung. (Abstract). Derm. Wschr. **82,** 2, 45—46, 1926.

GUNDERSEN, Edv. (1926b) Sop-sporecylindre i urinen i et tilfaelde av dysuria. Summary: "Mold casts" in the urine in a case of dysuria. Norsk Mag. Laegevidensk. **87,** 2, 124—129, 1926.

GUNDERSEN, Edv. (1926c) Svar til dr. philos. SOPP's bemerkninger til "Om sopsporecylindre i urinen". (Reply to Dr. Philos. SOPP's remarks on "Om sopsporecylindre i urinen"). Norsk Mag. Laegevidensk. **87,** 4, 344—346, 1926.

GUNDERSEN, Edv. (1934) Undersøkelser over patogene hudsoppers biologi med saerlig henblikk på variabilitet og pleomorfisme. (Investigations into the biology of pathogenic dermatomycetes with particular regard to their variability and pleomorphism). Avh. Norske Vidensk. Akad. Oslo. I. Mat. Naturv. Klasse 1934, Nr. 11. Pp. 44.

HADERUP, V. (1890) Om Invadering af patogene svampe fraa Mundhulen. (On the invasion of pathogenic fungi from the oral cavity. Lecture). Medicinsk Selskabs i København Forhandlinger 1890—91. 3 Møde, 11.11.1890. Hospitals-Tidende, 3 Raekke, **9,** 1, 17—18, 1891, and Bibliotek for Laeger, **83,** 7 Raekke, **2,** 90—91, 1891.

HADERUP, V. (1891) Om Selvinfektion fra Mundhulen. (On auto-infection from the oral cavity. Lecture). Skandinaviska Tandläkareföreningens Jubileumsfest i Göteborg 1891, Skand. Tandläkarefören. Tidskr. **1,** 54—67, 1892.

HAFSTRÖM, T. (1930) Fall av mycosis pulmonum. (Case of Mycosis pulmonum). Årsmöte, 1.6.1930. Svenska fören. för invärtes med. förhandl. 1930, pp. 203—214.

HAFSTRÖM, T. (1933) Ett fall av lungmykos. Diskussionsinlägg till DANIELSON, C. G.: Fall av pneumomykos. (A case of lung Mycosis. Same as in Sv. fören.f. invärtes medic. förhandl. 1930. Contribution to a discussion concerning DANIELSON's Fall av pneumomykos. Pp. 411—413). Sammankomst, 16.5.1933. Svenska Läkaresällsk. Förhandl. år 1933, pp. 389—414.

HAGEM, Oscar (1910) Einige Beobachtungen über die Verbreitung der Actinomyceten in der Natur (Vorläufige Mitteilung.). Annex (Chapter 7, pp. 197—205) to "Aktinomykosen (Straalesopsygdommen) i Norge," by HARBITZ, F. & BACKER GRØNDAHL, N., Videnskabs-Selskabets Skrifter. I. Math.-Naturv. Klasse, 1910, Nr. 7. (Christiania, 1910).

HALLBERG, V. (1938) See GULLBERG, Evert, 1938.

HALLBERG, V. (1940) En ny metod för tuberkelbacillfärgning. (A new method for staining Tuberculosis bacilli). Svenska Patologföreningen. Sammanträdet, 4.11. 1939. Nordisk Med. **5,** 1940: 1, 2, Nordisk Med. Tidskr. 164, 1940.

HALLDÉN, G. (1917) Fall av lungaktinomykos. (Case of Actinomycosis of the lung). Tuberkulosläkareföreningens förhandlingar. Sammanträde, 16.2.1916. Hygiea, **79,** 10, 484—491, 1917.

HANNOVER, Adolph (1839) Ueber eine contagiöse Confervenbildung auf dem Wassersalamander. Arch. Anat. Physiol wiss. Med. herausg. v. Dr. Johannes MÜLLER, **1839,** Jg., 338—347, 1839.

HANNOVER, Adolph (1842a) Om Entophyter paa Slemhinderne af det døde og levende menneskelige Legeme. (On Entophytes on the mucous membranes of the dead and living human organism). Ugeskrift for Laeger, **6,** 12, 177—190, 1842.

HANNOVER, Adolph (1842b) Ueber Entophyten auf den Schleimhäuten des todten und lebenden menschlichen Körpers. Arch. Anat. Physiol. wiss. Med. herausg. v. Dr. Johannes MÜLLER, **1842,** Jg., 281—295, 1842.

HANNOVER, Adolph (1847) F. T. BERG, Om Torsk hos Barn. Stockholm 1846. Kritisk Anmeldelse. (F. T. BERG, On Thrush in children. Stockholm 1846. A critical review). Ugeskrift for Laeger, 2 Raekke, **6,** 27, 417—431, 1847.

HANSEN, Emil Chr. (1879) Bidrag til Kundskab om hvilke Organismer, der kunne forekomme og leve i Øl og Ølurt. 1. Undersøgelser over de Organismer, som til forskjellige Tider af Aaret findes i Luften i og omkring Carlsberg, og som kunna udvikle sig i Ølurt. (Pp. 185—209). Contributions à la connaissance des organismes qui peuvent se trouver dans la bière et le moût de biere et y vivre. 1. Recherches sur les organismes qui, à différentes époques de l'année, se trouvent dans l'air, à Carlsberg et aux alentours, et qui peuvent se développer dans le moût de bière. (Appendix, pp. 49—67). Medd. Carlsberg Lab. **1,** 2 (1879), 185—209; Appendix, 49—67, 1876—1882.

HANSEN, Emil. Chr. (1882) Undersøgelser over de Organismer, som til forskjellige Tider af Aaret findes i Luften i og omkring Carlsberg, og som kunne udvikle sig i Ølurt. (Anden Meddelelse). (Pp. 381—454). Recherches sur les organismes qui, à différentes époques de l'année, se trouvent dans l'air, à Carlsberg et aux alentours, et qui peuvent se développer dans le moût de bière. (Deuxieme communication). (Appendix, pp. 197—218). Medd. Carlsberg Lab. **1,** 4, (1882), 381—454; Appendix, 197—218, 1876—1882.

HANSEN, Emil Chr. (1886) Om Analyser af Luftens Mikroorganismer. (On analyses of the microorganisms of the air. Lecture). Selskabet for Sundhedsplejen, 4.3. 1886. Hyg. Medd. 3 Raekke, **3,** 217—238, 1887.

HANSEN, Preben (1937) Familie med Favus. (Family with Favus). Dansk Derm. Selsk. Forhandl. Sept. 1936-Maj 1937, p. 10. Møde, 4.11.1937. Hospitalstidende, **80,** Bilaga, 1937.

HANSEN, Svend (1932) See BJERRUM, Oluf & HANSEN, Svend, 1932.

HARBITZ, Francis, (1898) Bidrag til laeren om actinomyces hominis. (Contribution to the principles of Actinomycosis hominis). Norsk Mag. Laegevidensk. **59,** 4 Raekke, **13,** 1—44, 1898.

HARBITZ, (Francis) (& GEIRSVOLD) (1904) Blastomyceter og blastomycoser. (Blastomycetes and Blastomycoses. Demonstration. Pp. 295—299). Forhandl. Med. Selsk. i Kristiania 1904, pp. 295—301. Møde, 26.10.1904. Norsk Mag. Laegevidensk. **65,** 5 Raekke, **2,** Bilaga, 1904.

HARBITZ, (Francis) (1909) Aktinomykose. (Actinomycosis. Case). Forhandl. Med. Selsk. i Kristiania 1909, pp. 215—218. Møde, 10.11.1909. Norsk Mag Laegevidensk. **70,** 5 Raekke, **7,** Bilaga, 1909.

HARBITZ, (Francis) (1910a) Aktinomykose. (Actinomycosis. Lecture). Forhandl. Selsk. i Kristiania 1910, p. 218. Møde, 7. 12.1910. Norsk Mag. Laegevidensk. **71,** 5 Raekke, **8,** Bilaga, 1910.

HARBITZ, Francis & BACKER GRØNDAHL, Nils (1910b) Aktinomykosen (Straalesopsygdommen) i Norge. Studier over dens forekomst, patologiske anatomi og bakteriologi. (Actinomycosis in Norway. Studies on its occurrence, pathological anatomy and bacteriology). Videnskabs-Selskabets Skrifter. I. Math.-Naturv. Klasse, 1910, Nr. 7. Pp. 211. (Christiania, 1910).

HARBITZ, Francis & BACKER GRØNDAHL, Nils (1911a) Om aktinomykosen i Norge. Studier over Sygdommens forekomst, dens patologiske anatomi og bakteriologi. Utdrag ur en afhandling, 1910. (On Actinomycosis in Norway. Studies on the occurrence of the disease, its pathological anatomy and bacteriology. Excerpt from a treatise, 1910). Norsk Mag. Laegevidensk. **72,** 5 Raekke, **9,** 6, 625—702, 1911.

HARBITZ, Francis & BACKER GRONDAHL, Nils (1911b) Die Strahlenpilzkrankheit (Aktinomykose) in Norwegen. Studien über ihr Vorkommen, ihre Lokalisation und Bakteriologie. Beitr. path. Anat. **50,** 193—243, 1911.

HARTMANN, Otto (1939) see BØE, J., HARTMANN, Otto & THJØTTA, Th., 1939.

HARTWIG, Chr. (1894) See FRIELE, J. & HARTWIG, Chr., 1894.

HASLUND, Otto (1911) Onychomycosis. Trichophyton capillitii, (Case). Dansk Dermatologisk Selskab. 84 Møde, 6.12.1911. Hospitalstidende, **55,** 5 Raekke, **5,** 23, 666—667, 1912.

HASLUND, Otto (1915) See BANG, H. & HASLUND, Otto, 1915.

HAXTHAUSEN, H. (1920) Sekundaere Trichophytider af forskellig Type hos 2 Søskende, begge inficerede med samme Svamp. (Secondary Trichophytics of different type in two siblings, both infected with the same fungus. Cases). Dansk Derm. Selsk. Forhandl. i Oktober 1919—Maj 1920, pp. 41—42. 146 Møde, 3.3. 1920. Hospitalstidende, **63,** Bilaga, 1920.

Haxthausen, H. (1928) Blastomycosis. (Cases). Dansk Derm. Selsk. Ferhandl Okt. 1928—Maj 1929, pp. 26—28. 214 Møde, 5.12. 1928. Hospitalstidende, **72,** Bilaga, 1929.

Haxthausen, H. (1929) Sekundaere Trikofytider lokaliserede til Hudpartier med perniotiske Karforandringer. (Secondary Trichophytids localized to skin regions with perniotic blood vessel changes. Case). Dansk Derm. Selsk. Forhandl. i September 1929—Maj 1930, pp. 12—13. 220 Møde, 2.10.1929. Hospitalstidende, **73,** Bilaga, 1930.

Haxthausen, H. (1932a) Aktinomykose. Jodbehandling. Erysipelas. (Actinomycosis. Iodine treatment. Erysipelas. Case). Dansk Derm. Selsk. Forhandl. Sept. 1931—Maj 1932, p. 33.240 Møde, 3.2.1932. Hospitalstidende, **75,** Bilaga, 1932.

Haxthausen, H. (1932b) Udbredt Moniliainfektion af Huden og Mundslimhinder med ejendommelig Neglelidelse og sikundaert Exanthem. (Disseminated Monilia infection of the skin and oral mucous membranes, with a peculiar affection of the nails and secondary rash. Case). Dansk Derm. Selsk. Forhandl. Sept. 1932—Maj 1933, p.4. 244 Møde, 5.10. 1932. Hospitalstidende, **76,** Bilaga, 1933.

Haxthausen, H. (1935) Demonstration of cases. Monilia infection. (P. 598). Proc. IX. Meet. Northern Derm. Soc., Copenhagen 10—12 June, 1935. Acta derm.—vener. **17,** 187—647, 1937.

Haxthausen, H. (1936) 2 Tilfaelde af Favus, behandlet med Paraoxybenzoesyreaethylester. (Two cases of Favus treated with Ethyl ester of paraoxybenzoic acid). Dansk Derm. Selsk. Forhandl. Sept. 1936—Maj 1937, p. 38. 273 Møde, 4.3.1936. Hospitalstidende, **79,** Bilaga, 1936.

Haxthausen, H. (1939) Fodsvamp. (Foot Mycosis. Lecture). Ugeskrift for Laeger, **101,** 14, 410—413, 1939.

Hedenius, P. (1873) Om några parasitsvampar och deras betydelse för infectionsläran. (On some parasitic fungi and their significance for the principles of infection Lecture). 22.11.1872. Upsala Läkarefören. Förhandl. **8,** 3, 266—294, 1872—1873.

Heiberg, Hjalmar (1887) Orienterende Oversigt over Laeren om de pathogene Bakterierne hos Mennesket. (An introductory survey on the principles of pathogenic bacteria in man). Klinisk Aarbog. Medd. Rigshospitalet og Fødselstiftelsen i Kristiania, **4,** 152—188, 1887.

Helleberg, G. (1938) See Gullberg, Evert, 1938.

Hellens, v., O. (1902) Till kännedomen om genom Aspergillus fumigatus framkallade förändringar i lungerna. (To the knowledge of changes in the lungs caused by Aspergillus fumigatus). Förhandlingar vid Nordiska Naturforskare- och Läkaremötet i Helsingfors, 7—12 Juli 1902. C. R. Congr. Natur. Méd. du Nord tenu à Helsingfors, 7—12 Juillet 1902. 9. Sektionen för patologisk anatomi och bakteriologi, pp. 43—44.

Hellens, v., O. (1905) Zur Kenntnis der durch den Aspergillus fumigatus in den Lungen hervorgerufenen Veränderungen. Arb. Path. Inst. Univ. Helsingfors (Finland), **1,** 1—2, 313—343, 1905.

Hellerström, (Sven) (1934) Fall von provozierten Mykiden. Verh. Derm. Ges. zu Stockholm. Sitzung, 11.4.1934. Acta derm.-vener. **16,** 1, 106—107, 1935.

Hellerström, S(ven) (1940a) Akute Dermatitiden entstanden bei der Zubereitung essbarer Pilze. (Publ. in Acta derm.-vener. **22,** 331, 1941). Verh. Derm. Ges. in Stockholm 1940. Sitzung, 9.10.1940. Acta derm.-vener. **23,** 1, 65—66, 1942/43.

Hellerström, Sven (1940b) Sensitization to edible mushrooms. Acta derm.-vener. **22,** 4, 331—336, 1941.

Hellström, Georg (1895) En undersökning angående aktinomykosens förekomst hos menniskan i Sverige. (An investigation concerning the occurrence of Actinomycosis in man in Sweden). Sjunde Allmänna Svenska Läkaremötet 1895. Hygiea, **57,** 9, 362—365, 1895.

Hempel, C. (Editor) (1849) Om parasitiske Planter som forekomme paa den menneskelige Organisme. (On parasitic plants occurring in the human organism). Ugeskrift for Laeger, 2 Raekke, **11,** 18, 273—283, 1849.

Hempel and Engelstad (Editors) (1855) On Herpes tonsurans (Cazenavi). (On Herpes tonsurans Cazenavi). Ugeskrift for Laeger, 2 Raekke, **22,** **3,** 33—45 1855.

HEMPEL and ENGELSTAD (Editors) (1859) Dermatologiske Skizzer. 3. Bazins Oppfattelse og Behandling af de parasitiske Affektioner i Huden. (Dermatological sketches. 3. BAZIN'S conception and treatment of parasitic skin affections). Ugeskrift for Laeger, 2 Raekke, **31**, 28, 437—448; 29, 453—464; 30, 469—476, 1859.

HENSCHEN, Folke (1930) See BERGMAN, R. & HENSCHEN, Folke, 1930.

HERLOFSEN, Th. (1897) Et tilfaelde av actinomycosis hominis. (A case of Actinomycosis hominis). Norsk Mag. Laegevidensk. **58,** 4 Raekke, **12,** 1141—1143, 1897.

HERMANSSON, K. A. (1936) Herpes hos häst som orsak till hudsjukdomar hos människor. Summary: Herpes in horse as the cause of skin-disease in man. Skand. Vet. Tidskr., **26,** 27—33, 1936.

HERTZ, Rolf (1909) Et Tilfaelde af Actinomycosis pleurae. (A case of Actinomycosis pleurae). Dansk Paediatrisk Selskabs Forhandlinger. 8 Møde, 9.12.1909. Nordisk Tidsskr. Terapi **8,** 264—267, 1909—1910.

HERTZ, Rolf (1935) Et Tilfaelde af Lymfosarcoma abdominale og et Tilfaelde af Actinomycosis pleurae fistulosa. (A case of Lymphosarcoma abdominale and a case of Actinomycosis pleurae fistulosa). Dansk Paediatrisk Selskabs Forhand linger. 159 Møde, 22.4.1935. Ugeskrift for Laeger, **98,** 3, 55—56, 1936.

HEYERDAHL, (S.A.) (1914a) Et tilfaelde af ansigtsaktinomykose. (A case of Actinomycosis of the face. To be publ. in Nordisk Tidsskrift for Terapi, 1914). Forhandl. Med. Selsk, i Kristiania 1914, p. 3. Møte, 14.1.1914. Norsk Mag. Laegevidensk. **75,** 5 Raekke, **12,** Bilaga, 1914.

HEYERDAHL, S.A. (1914b) Et tilfaelde af aktinomykose behandlet med radium. (A case of Actinomycosis treated with Radium). Nordisk Tidsskr. Terapi, **12,** 169—173, 1913—1914.

HEYERDAHL, S. A. (1915) 4 tilfaelder av aktinomykose behandlet med radium. (Four cases of Actinomycosis treated with Radium). Forhandl. Med. Selsk. i Kristiania 1915, pp. 175—179. Møte, 24.11.1915. Norsk Mag. Laegevidensk., **76,** 5 Raekke, **13,** Bilaga, 1915.

HEYERDAHL, S. A. (1916) Nogle tilfaelder af Aktinomykose helbredet med radium. (Some cases of Actinomycosis healed with Radium). Förhandl. Nordisk Kirurg. Fören. 11: te möte i Göteborg, 6—8 Juli 1916, pp. 110—114.

HEYERDAHL, S. A. (1919) Actinomycosis treated with Radium. J. Amer. med. Ass. **73,** 26, 1928—1929, 1919.

HEYERDAHL, S. A. (1925) Actinomycosis of the Face and Neck Treated with Radium. Read before the First International Congress of Radiology, London,1925. Brit. J. Radiol. (B. J. R. Section), **31,** 306, 1—14, 1926.

HEYERDAHL, S. A. (1927) Über die Radiumbehandlung der Aktinomykose des Gesichts und des Halses. Strahlenther. **25,** 4, 679—691, 1927.

HINRICSSON, Harry (1940) See PALM, B. & HINRICSSON, Harry, 1940.

HIORTH, Wilh. (1887) Nogle Undersøgelser om parasitaere Haarsygdomme, saerlig Skurv, paa Kristiania Almuskoler. (Some investigations into parasitic hair diseases, especially Favus, in the Elementary schools in Kristiania). Norsk Mag. Laegevidensk., **48,** 4 Raekke, **2,** 500—514, 1887.

HIORTH, Wilh. (1888) Om Forekomsten af Favus i Norge. (On the occurrence of Favus in Norway). Norsk Mag. Laegevidensk., **49,** 4 Raekke, **3,** 105—141; 177—187, 1888.

HJELMMAN, J. W. (1914) Sädesienitaudi Kauvatsalla ja Kiikoisissa. (Actinomycosis in Kauvatsa and Kiikoinen). Satakunta, 1914. Quoted by BROFELDT, Simo, 1926.

HJELT (1864a) Om vaginal-katarrher hos quinnan, förorsakade af alger och svampar. (On vaginal inflammations in women, caused by algae and fungi. Lecture). Finska Läkaresällskapets sammanträde, 19.2.1864. Notisblad för Läkare och Pharmaceuter, 1864, p. 104. Finska Läkare-Sällsk., Handl. **1864,** Bihang, 1864.

HJELT (1864b) Svampar såsom ursak till inflammation i vagina. Meddeledse. Utdrag ur: Årsberättelse för 1864. (Fungi as cause for vaginal inflammation. Communication. Excerpt from the annual report for 1864. p. 1197). Finska Läkare-Sällsk. Handl., **9,** 1189—1214, 1866.

HÖK (1852) Fusarium heterosporium — demonstration. (Demonstration. See also LEVIN, and WAHLBERG, both 1852). Förhandlingar vid Svenska Läkare-Sällskapets sammankomster 1851—1852. Sammankomst, 18.5.1852. Hygiea, **14**, 12, 752, 1852.

HØYGÅRD, (Arne), jr. (1934a) To tilfeller av abdominal aktinomykose utgående fra appendix. (Two cases of abdominal Actinomycosis originating from the appendix. Publ. in Medic. Revue, **52**, 412, 1935). Det Medicinske Selskab i Bergen. Møte, 6.12.1934. Medicinsk Rev. **52**, 4, 187—188, 1935.

HØYGÅRD, Arne, jr. (1934b) To tilfeller av abdominal aktinomykos utgående fra appendix. (Two cases of abdominal Actinomycosis originating from the appendex). Medicinsk Rev. **52**, 9, 412—418, 1935.

HOLLAND, Wilh. (1913) Et tilfaelde av primaer hudaktinomykose med nogen bemerkninger om aktinomykosens inkubation og latens. (A case of primary skin Actinomycosis, with some remarks on the incubation and latency of Actinomycosis). Norsk Mag. Laegevidensk. **74**, 5 Raekke, **11**, 1048—1075, 1913.

HOLM, Per (1930) Comparative Studies on Some Pathogenic Anaerobic Actinomyces. (Preliminary report). Liber Gratulatorius in Honorem Thorvald MADSEN 18 Februaris 1930. (Communications de l'Institut Sérothérapique de l'Etat Danois, **20**). Acta path. microbiol. Scand. Supplementum 3, pp. 151—156, 1930.

HOLM, Per (1932) Quoted by BJERRUM, O. & HANSEN, S., 1932.

HOLST, Axel (1887) See CONRADI, 1887.

HOLST, Axel (1888) Et Tilfaelde af Actinomycosis hominis. (A case of Actinomycosis hominis). Norsk Mag. Laegevidensk. **49**, 326—328, 1888.

HOLST, Axel (1890) Oversigt over Bakteriologien for Laeger og Studerende. Smittestoffer af mugsoppens gruppe. Smittestoffe af gjaersoppens gruppe. Aktinomykosis. (Survey of bacteriology for physicians and students. Contagiums of the mould fungus group, pp. 99—101. Contagiums of the yeast fungus group, p. 102. Actinomycosis, pp. 168—171). H. Aschehoug & Co, Kristiania, 1890. Pp. 192.

HOLST, Axel (1901) Oversigt over Bakteriologien for Laeger og Studerende. 2den omarbeidede Udgave. Aktinomykosis. Smittestoffer af soppenes gruppe. (Survey of bacteriology for physicians and students. Second revised edition. Actinomycosis, pp. 206—210. Contagiums of the fungus group, pp. 210—212). H. Aschehoug & Co (W. Nygaard), Kristiania, 1901. Pp. 233.

HOLST, Peter F. (1895) Aktinomykosis hominis. (Case). Forhandlinger i det Medicinske Selskab i Kristiania 1895, pp. 4—6. Møde, 9.1.1895. Norsk Mag. Laegevidensk. **56**, 4 Raekke, **10**, Bilaga, 1895.

HOLST, Peter F. (1897) Lungeaktinomykose. (Lung Actinomycosis. Cases). Forhandl. Med. Selsk. i Kristiania 1897, pp. 194—198. Møde, 15.9.1897. Norsk Mag. Laegevidensk. **58**, 4 Raekke, **12**, Bilaga, 1897.

HOMÉN (1892) Förevisning af preparat af ett fall af bukaktinomykos. (Demonstration of preparations from a case of abdominal Actinomycosis). Finska Läkaresällskapets sammanträde, 17.12.1892. Finska Läkaresällsk. Handl. **35**, 2, 150, 1893.

HOMÉN (1893) See KROGIUS, 1893.

HÜBERT, K. (1937) Favus pityriasiforme. (Case). Verh. Norw. Derm. Ges. Sitzung, 9.12.1937. Acta derm.—vener. **20**, 3, 330, 1939.

HUSS, R. (1938) See GULLBERG, Evert, 1938.

ILMONI, J. (1842) Nosographiska Anmärkningar om Diabetes Mellitus. (Nosographical remarks on Diabetes mellitus). Förhandl. Skand. Naturforsk. tredje Møte i Stockholm, 13—19 Juli, 1842, pp. 840—847.

JÄDERHOLM (1871) See LILJENROTH, A., 1871.

JENSEN, C. E. & KIØRBOE, Aage (1910) Sporotrikose. En Oversigt. (Sporotrichosis. A survey). Ugeskrift for Laeger, **72**, 11, 305—315, 1910.

JENSEN, H. L. (1931) Contribution to our Knowledge of the Actinomycetales. II. The Definition and Subdivision of the Genus Actinomyces, with a Prelinimary Account of Australian Soil Actinomycetes. Proc. Linn. Soc. N. S. W. **56**, Part 4, 345—370, 1931.

JENSEN, Rich. (1883) Straalesvampsygdom hos Kvaeget. (Actinomycosis in Cattle). Kgl. Landhusholningsselskab, 28.2.1883. Tidsskr. Landøkonomi, Kjøbenhavn, 5 Raekke, **2**, 215—236, 1883.

JENSEN, Vilhelm P. H. (1902) Histogenesis of the nodules produced by subcutaneous injection of Saccharomyces neoformans Sanf. Festskrift ved Indvielsen af Statens Serum Institut 1912. Contribution from the University Laboratory for

Medical Bacteriology to Celebrate the Inauguration of the State Serum Institute. Copenhagen, 1902, 10, Pp. 19.

Jensen, Vilh(elm) P. H. (1903) Undersøgelser over Patogen Gaer. (Investigations into pathogenic yeasts). Thesis, Copenhagen, 1903. Det Nordiske Forlag, Ernst Bojesen, København, 1903. Pp. 128.

Jersild (1909) Trikofyti, Röntgenbehandling, (Trichophytosis, X—ray treatment). Dansk Dermatologisk Selskab. 67 Møde, 5.5.1909. Hospitalstidende, **52,** 5 Raekke, **2,** 33, 1044, 1909.

Jersild, O. (1925) Demonstration af to Søskende med Trichophytia. (Demonstration of two siblings with Trichophytosis). Dansk Derm. Selsk. Forhandl. 1924—1925, p. 25. 187 møde, 4.3.1925. Hospitalstidende, **68,** Bilaga, 1925.

Jersild, O. (1934) Cu-Jontoforese af Epidermofytier. (Copper iontophoresis in Epidermophytoses. To be published later). Dansk Derm. Selsk. Forhandl. Sept. 1933-Maj 1934, p. 30. 255 Møde, 7.2.1934. Hospitalstidende, **77,** Bilaga, 1934.

Jersild, O. & Plesner, Marie (1936) Traitement de l'épidermophytie des extrémités par iontophorèse de cuivre. Société Française de Dermatologie et de Syphiligraphie. Séance, 13.2.1936. Bull. Soc. Franç. Derm. Syphiligr. **43,** 2, 450—457, 1936.

Jersild, O. & Plesner, Marie (1939) Behandling af Fodsvamp med Kobber-iontoforese. (Treatment of foot Mycosis with copper iontophoresis. Same as Acta derm. -vener. **21,** 268, 1940). Ugeskrift for Laeger, **101,** 50, 1475—1479, 1939.

Jersild, O. & Plesner, Marie (1940) Treatment of epidermophytia on the extremities with iontophoresis of copper. Acta derm.-vener. **21,** 2, 268—279, 1940.

Jervell, Kr. (1895) Fire Tilfaelde af Aktinomykose, udgaaende fra Mundhulen. (Four cases of Actinomycosis originating from the oral cavity). Festskrift Professor Hjalmar Heiberg, pp. 100—112. A. W. Brøggers Bogtrykkeri, Kristiania, 1895.

Johan-Olsen (Sopp), Olav. See also Sopp (Johan-Olsen), Olav.

Johan-Olsen, O(lav) (1886a) Hudsygdom, frembragt ved en Mugsop, voxende i en Lister's Bandage. (Skin disease caused by a mould fungus, growing in a Lister's bandage). Norsk Mag. Laegevidensk. **47,** 4 Raekke, **1,** 244—248, 1886.

Johan-Olsen, O(lav) and Gade, F. G. (1886b) Undersögelser over Aspergillus subfuscus som patogen mugsop. (Investigations into Aspergillus subfuscus as a pathogenic mould fungus). Nordiskt Med. Ark. **18,** 9, 1—52, 1886.

Johan-Olsen, Olav (1889) Fremvisning af Kulturer af Herpes tonsurans. (Demonstration of cultures of Herpes tonsurans). Forhandl. Norske Med. Selsk. i Kristiania 1889. Møde, 13.2.1889. Norsk Mag. Laegevidensk. **50,** 4 Raekke, **4,** Bilaga, 1889.

Johan-Olsen, Olav (1893) Om sop paa levende jordbund. (On fungi on living media. Thesis, Kristiania, 1893. Hjalmar Biglers Forlag, Kristiania, 1893. Pp. 136.

Johan-Olsen, Olav (1897) Zur Pleomorphismusfrage. Cbl. Bakt. Parasitenk. 2 Abt. **3,** 11/12, 273—284, 1897.

Johan-Olsen Sopp, Olav (1915) Mikrobernes sygdomme. Infektionsbegreppets utvidelse. Et forløbig resumé. English Summary: The diseases of the microbes. (Widening the conception of infection. A preliminary summing up). Norsk Mag. Laegevidensk. **76,** 5 Raekke, **13,** 1073—1099. 1915.

Johansen, Anders (1893) Kerion Celsi. Hospitalstidende, **36,** 4 Raekke, **1,** 36, 917—918, 1893.

Kaarsberg, F. (1887) Kasuistiske Meddelelser fra Kommunehospitalets Afd. I. 3. Aktinomykosis. (Casuistical communications from the First Department of the Municipal Hospital. 3. Actinomycosis. Pp. 7—11). Nordiskt Med. Ark. **19,** 22, 1—20, 1887.

Kaarsberg, J. (1914) Tumor parietis abdominis. (Case). Foreningen for Gynaekologi og Obstetrik. 87 Møde, 18.3.1914. Ugeskrift for Laeger, **76,** 44, 1917—1919, 1914.

Kanerva, Aimo (1931) Keratitis aspergillina. Deutsches Referat: Ein Fall von Keratitis aspergillina. Duodecim, **47,** 11, 1004—1008, 1931

Kanerva, Aimo (1932) Ein Fall von Keratitis aspergillina. Acta ophthal., Kbh., **10,** 376—381, 1932.

Kiaer (1887) Berättelse. (Report on Herpes tonsurans in children, with a simultaneous occurrence of the disease in cattle on the same farm). Forhandl. Norske Med. Selsk. i Kristiania 1887, p. 4. Møde, 12.1.1887. Norsk Mag. Laegevidensk. **48,** 4 Raekke, **2,** Bilaga, 1887.

KIØRBOE, Aage (1910) See JENSEN, C. E. & KIØRBOE, Aage, 1910.

KISSMEYER, A. (1914) See RASCH, C. & KISSMEYER, A., 1914.

KISSMEYER, (A.) (1919) Trichophyton rosaceum — Kultur. (Culture demonstration). Dansk Dermatologisk Selskab. 137 Møde, 2.4.1919. Hospitalstidende, **62,** 30, 896, 1919.

KISSMEYER, (A) (1934) Sekundaere Trichophytider opstaaet i Tilstluning til Trichophytin-Intradermoreaktion. (Secondary Trichophytids occurring in connection with an intracutaneous Trichophytin reaction). Dansk Derm. Selsk. Forhandl. September 1933—Maj 1934, pp. 31—32. 255 Møde, 7.2.1934. Hospitalstidende, **77,** Bilaga, 1934.

KISSMEYER, A. (1938) Om Behandling af Epidermomykoser af Haender og Fødder med Jontophorese a.m. Jersild. (On the treatment of Epidermomycoses of the hands and feet with iontophoresis according to JERSILD). Ugeskrift for Laeger, **100,** 17, 439—440, 1938.

KJERSCHOW (1892) See RAMM, 1892.

KNUTSON, D. (1933) Ett fall av aktinomycos. pulmon. (A case of Actinomycos pulmonum). Tuberkulosläkareförenings Förhandlingar 1933. Sammanträde, 8.3. 1933. Hygiea, 96, 17, 612—613, 1934.

KÖSTER, H. (1893) Primär actinomycosis pulmonum. (Primary Actinomycosis pulmonum). Göteborgs Läkaresällsk. Förhandl. 1893, 2 häftet, pp. 181—186.

KÖSTER, H. (1894) Primär actinomycosis pulmonum med metastaser. (Primary Actinomycosis pulmonum with metastases). Göteborgs Läkaresällsk. Förhandl. 1894, 1 häftet, pp. 1—9.

KRISTENSON, Anders (1940) Några erfarenheter med färgning av tuberkelbaciller ad modum Hallberg. (Some experiences with the staining of Tuberculosis bacilli according to HALLBERG's method). Svenska Läkartidningen, **37,** 20, 892—896, 1940.

KROGIUS (1893) Ett fall af akut förlöpande aktinomykos. Med demonstration av sektionsfynd av HOMÉN. (A case of Actinomycosis running an acute course. With a demonstration of the post-mortem finds by HOMÉN). Finska Läkaresällskapets sammanträde, 18.3.1893. Finska Läkaresällsk. Handl. **35,** 5, 388, 1893.

KROGIUS, Ali (1926a) Ett fall av aktinomykos i tibia. (A case of Actinomycosis of the tibia). Möte, 15.5.1926. Finska Läkaresällsk. Hand.l **68,** 6, 597, 1926.

KROGIUS, Ali (1926b) Till kännedomen om den hematogena aktinomukosen i de långa rörbenen. Deutsches Referat: Zur Kenntnis der hämatogenen Aktinomykose der langen Röhrenknochen. Finska. Läkaresällsk. Handl. **69,** **1,** 1—**11,** 1926.

KROGIUS, Ali (1928) Zur Kenntnis der hämatogenen Aktinomykose der langen Röhrenknochen. Acta chir. Scand. **63,** 121—132, 1928.

LAACHE, S. (1892) Kliniske Meddelelser fra Rigshospitalets medicinske Afdelning B. 7. Aktinomycosis hominis intestinalis. (Clinical communications from the Department of Medicine B at the Rigs Hospital. 7. Actinomycosis hominis intestinalis). Norsk Mag. Laegevidensk. **53,** 4 Raekke, **7,** 12, 1435—1450, 1892.

LAACHE, S. (1906) Zur Kasuistik des doppelseitigen Empyems. Berl. klin. Wschr. **43,** 3, 65—67, 1906.

LANGENBECK, B. (1839) Auffindung von Pilzen auf der Schleimhaut der Speiseröhre einer Typhus-Leiche. Neue Not. Nat. Heilk. **12,** 10 (252), Spalten 145 —147, 1839.

LANGENSKIÖLD, F. (1916) Ett fall av kroniskt förlöpande, varig mastit, förorsakad av en cladotrhixart. (A case of purulent Mastitis with a chronic course, caused by a Cladothrix species). Finska Läkaresällsk. Handl. **58;** 2, 10, 1538—1551, 1916.

LANGENSKIÖLD, F. (1926) Über die primäre Aktinomykose der Brustdrüse. Acta chir. Scand. **59,** 23—26, 1925/26.

LARSEN (1844) Cliniske Forelaesninger. Tredje Forelaesning. (Clinical lectures. Third lecture). Ugeskrift for Laeger, 2 Raekke, **1,** 7, 104—109, 1844.

LECZINSKY, C. G. (1938) See GULLBERG, Evert, 1938.

LEHMANN, Jørgen & NIELSEN, Holger E. (1939a) Et Tilfaelde af Beri-Beri (efterfulgt af Pellagra), verificeret og fulgt under Bedringen gennem B_1-Vitaminanalyser paa Blodet. (A case of Beri-Beri, succeeded by Pellagra, verified and followed up during the recovery with analyses of B_1-Vitamine in the blood). Nordisk Med. **1,** 1939: 1, 5, Hospitalstidende, 289—292, 1939.

Lehmann, Jørgen & Nielsen, Holger (E.) (1939b) B_1-Vitaminbestemmelse i Blod med Schopfers Svampmetode. Autoreferat. (B_1 determination in the Blood by Schopper's fungus-method. Abstract by the authors). Nordiske Kongress for indre medicin i Oslo, 27—29 juni 1939. Nordisk Med. **2,** 1939: 2, 25, Nordisk Med. Tidskr. 1943, 1939.

Lennander, K. G. (1890) Ett fall af actinomyces linguae. (A case of Actinomycosis linguae). Foreningens särskilda angelägenheter, p. 5. 24.10.1890. Upsala Läkareforen. förhandl. **26,** Bilaga, 1890—1891.

Lennmalm, F. (1911) Per Henrik Malmsten. Minnesteckning. (Biography). Svenska Läkaresällsk. Handl. **39,** 1—99, 1911.

Levin, P. A. (1852a) Om en i Nerke nu gängse endemisk sjukdom uppkommen genom den osunda beskaffenheten af förra årets rågskörd. (On an endemic disease at present current in Nerke, and caused by the unwholesome state of last year's rye-crop. See also Wahlberg, and Hök). Svenska Läkare-Sällskapet, 13.1.1852. Hygiea, **14,** 2, 76—82, 1852.

Levin, P. A. (1852b) Sjukdom i Nerke efter förtärandet af ny råg. (A disease in Nerke due to the consumption of fresh rye). Förhandlingar vid Svenska Läkare-Sällskapets sammankomster 1851—1852. Sammankomst, 13.1.1852. Hygiea, **14,** 5, 308—309, 1852.

Liljenroth, A. (1871) Sex fall av Aspergillus flavescens i yttre hörselgången. (Six cases of Aspergillus flavescens in the external auditory canal. Demonstration of microscopical slides by Jäderholm). Förhandlingar vid Svenska Läkare-Sällskapets sammankomster år 1871, pp. 248—249. Sammankomst, 21.11.1871. Hygiea, **33,** Bilaga, 1871.

Liljenroth, A. (1872) Om otomykosis. (On Otomycosis). Nordiskt Med. Ark., **4,** 13, 1—15, 1872.

Lindegaard Nielsen, O. (1931) Et Tilfaelde af primaer Lungaktinomykos. (A case of primary lung Actinomycosis). Medicinsk Selskab for Fyens Stifts Forhandlinger 1930—1931, pp. 50—54. 35 Møde, 25.1.1931. Hospitalstidende, **74,** Bilaga, 1931.

Lindgren, Jorma (1931) Muutamia harvinaisia aktinomykoosin lokalisaatiomuotoja. Deutsches Referat: Einige ungewöhnliche Lokalisationsformen der Aktinomykose. Duodecim, **47,** 12, 1109—1121, 1931.

Lindqvist, C. (1867) Quoted by Fries, O. R., 1867.

Ljunggren, C. A. (1895) Om Actinomycosis hominis och dess förekomst inom Södra Skåne. (On Actinomycosis hominis and its occurrence in Southern Scania). Nordiskt Med. Ark. **27,** 27, 1—26, 1895.

Lodberg, C. V. (1911) Et Tilfaelde af Svampkonkrement i Canaliculus lacrymalis inf. med Praeparatdemonstration. (A case of fungus concretion within the Canaliculus lacrimalis inferior with demonstration of preparation). Det Oftalmologiske Selskab i København. 49 Møde, 15.2.1911. Hospitalstidende, 5 Raekke, **4,** 45, 1289—1290, 1911.

Løvdal, Arne (1937) Et tilfelle av aktinomykose opstått efter tannekstraksjon. (A case of Actinomycosis due to tooth extraction). Norske Tannlaegeforen. Tidende, **47,** 5, 224—230, 1937.

Lomholt, S(vend) (1931) Epidermofytia pedum med sekundaere Epidermofytider paa Haender og Arme. (Epidermophytia pedum with scondary Epidermophytids on the hands and arms). Dansk Derm. Selsk. Forhandl. Sept. 1931.-Maj 1932, p. 21. 239 Møde, 2.12.1931. Hospitalstidende, **75,** Bilaga, 1932.

Lomholt, Svend (1932a) To Tilfaelde af Epidermofyti paa Fødderne med haematogen Utsaed af Epidermofytider paa Haender, Arme og Ben. (Two cases of Epidermophytosis of the feet with a haematogenous dissemination of Epidermophytids on hands, arms, and legs). Hospitalstidende, **75,** 1, 75—83, 1932.

Lomholt, S(vend) (1932b) Epidermophyti paa venstre Fod med udtalt dyshidrosiforme Epidermophytider paa Haender og Underarme. (Epidermophytosis of the left foot with pronounced dyshidrosiform Epidermophytids on the hands and forearms. Case). Dansk Derm. Selsk. Forhandl. Sept. 1931-Maj 1932, p. 44-45. 241 Møde, 2.3.1932. Hospitalstidende, **75,** Bilaga, 1932.

LOMHOLT, S(vend) (1932c) Leukonykia epidermophytica. (Case). Dansk Derm. Selsk. Forhandl. Sept. 1931-Maj 1932, pp44—45. 241 Møde, 2.3.1932. Hospitalstidende, **75**, Bilaga, 1932.

LOMHOLT, Svend (1932d) Universelle, dyshidrosiforme og erythematøse Trichofytider paa Krop og Ekstremiteter efter Trichofyti paa v. Hand. (General dyshidrosiform and erythematous Trichophytids of the body and extremities in consequence of Trichophytosis of the left hand. Case). Dansk Derm. Selsk. Forhandl. Sept. 1931-Maj 1932, pp. 45—47. Møde, 2.3.1932. Hospitalstidende, **75**,, Bilaga, 1932.

LOMHOLT, S(vend) (1932e) Eczema of the Feet Caused by the Epidermophyton interdigitale, Illustrated by an Investigation on 97 Students. Proc. VIII. Meeting Northern Derm. Soc. Stockholm 2—4 June, 1932. Acta derm.-vener. **13,** 5, 432—442, 1932.

LOMHOLT, S(vend) (1933) Eczem paa Fødderne, fremkaldt af Epidermophyton interdigitale, belyst ved en Undersøgelse over 97 Studenter. (Eczema of the feet caused by the Epidermophyton interdigitale, illustrated by an investigation on 97 students. Same as Acta derm.-vener. **13,** 432, 1932). Ugeskrift for Laeger, **95,** 4,97—101, 1933.

LOMHOLT, Sv(end) (1934a) Et nyt Antimycoticum. (Offentliggjort andensteds). (A new antimycotic. Published elswhere). Dansk Derm. Selsk. Forhandl. Sept. 1933-Maj 1934, pp. 61—62. 258 Møde, 2.5.1934. Hospitalstidende, **77,** Bilaga, 1934.

LOMHOLT, Svend (1934b) Et nyt Middel til behandlingen af mykotisk Ekzem paa Fødderne. (A new remedy for the treatment of mycotic Eczema of the feet). Ugeskrift for Laeger, **96,** 25, 655—657, 1934.

LOMHOLT, Svend (1934c) Profylaktiske Foranstaltninger mod Svampeczem paa Fødderne. (Prophylactical measures against fungous Eczema of the feet). Nordisk Med. Tidskr. **8,** 41, 1362—1363, 1934.

LOMHOLT Svend (1936) Forekomsten af Vorter og Taasvamp hos Skolebørn i København, paa Grundlag af en Undersøgelse af 1400 Børn. (Occurrence of warts and Mycoses of the feet in school children in Copenhagen, on the basis of an examination of 1400 children). Ugeskrift for Laeger, **98,** 35, 799—801, 1936.

LOMHOLT, Svend (1938a) Über die Fussmykosen in Kopenhagen und ihre Behandlung mit Mykokten. Med. Klin. **34,** 4, 118—119, 1938.

LOMHOLT, Svend (1938b) Om de almindeligaste Svampelidelser og deres Behandling. (On the most common mycotical disorders and their treatment). Maanedsskr. prakt. Laegegerning og social Med. **16,** 6, 222—236, 1938.

LOMHOLT, Svend (1939) Hudsygdommene og deres Behandling. Kortfattet Fremstilling for den praktiserende Laege. XV. Neglelidelser. XIX. Dermatomykoser. (Skin diseases and their treatment. A short presentation for the practitioner. XV. Nail affections. Mycotic nail affections, pp. 174—175. XIX. Dermatomycoses pp. 217—240). G.E.C. Gads Forlag, København, 1939. pp. 327.

LOMHOLT, Svend (1940) Kogesprit til Strømpedesinfektion ved Taasvamp. (Methylated spirits for the desinfection of stockings in case of foot Mycosis). Ugeskrift for Laeger, **102,** 30, 788, 1940.

LUND, Aage (1921) Über die Wirkung des Follikelhormones auf das Wachstum einiger Mikroorganismen. C. R. Lab. Carlsberg, Série phys. **21,** 12, 231—238, 1921.

LUNDSGAARD, K. K. K. (1899) Et Tilfaelde af Hypopyonkeratitis med Renkultur af Gaer. (A case of Hypopyon keratitis yielding a pure culture of yeast). Hospitalstidende, **42,** 4 Raekke, **7,** 41, 971—975, 1899.

LUNDSGAARD, K. K. K. (1900) Ein Fall von Hypopyonkeratitis mit Reinkultur von Hefe. Klin. Mbl. Augenheilk. **38,** 13—19, 1900.

MAGNUS, v., Rigmor (1931) Untersuchungen über einen unter gewissen Wachstumsbedingungen kapsulierten Aktinomyces. Zbl. Bakt. Parasitenk. I. Abt., Originale, **120,** 1/2, 113—116, 1931.

MAGNUS, v., Rigmor (1936) Undersøgelser over en Gruppe Actinomyceter isolerede fra Menneskets Svalg. English Summary. (Investigations into a group of Actinomycetes isolated from human throat). Thesis, Copenhagen, 1936. Levin & Munksgaard, Ejnar Munksgaard, København, 1936 Pp. 132.

Magnusson, Hilding (1928) The Commonest Forms of Actinomycosis in Domestic Animals and their Etiology. Acta path. microbiol. Scand. **5**, 170—245, 1928.

Malm (1893) Jodkaliumbehandlingen af Aktinomykose. (Potassium iodide treatment of Actinomycosis). Forhandl. Med. Selsk. i Kristiania i Aaret 1893, p.83. Møde, 8.3.1893. Norsk Mag. Laegevidensk. **54,** 4 Raekke, **8,** Bilaga, 1893.

Malmsten (P.H.) (1844a) Ett fall af så kallad Teigne tondante. Diskussionsinlägg. (A case of so-called Teigne tondante. Contribution to a discussion. To be dealt with in greater detail later on). Svenska Läkare-Sällskapets sammankomst, 7.5.1844. Förhandl. Svenska Läkare-Sällsk. sammankomster från och med Octob. 1843 till och med Sept. 1844, p. 62—63.

Malmsten, (P.H.) (1844b) Teigne tondante. (Report on the case mentioned in 7.5.1844). Svenska Läkare-Sällskapets sammankomst, 2.7.1844. Förhandl. Svenska Läkare-Sällsk. sammankomster från och med Octob. 1843 till och med Sept. 1844, p. 70.

Malmsten, P. H. (1844c) Den sjukdoms vegetabiliska natur, som Mahon beskrifvit under namn af Teigne tondante (Squarus tondens) och som man i allmänhet ansett vara identisk med Willans Porrigo decalvans. (The vegetable nature of the disease which Mahon described under the name of Teigne tondante (Squarus tondens) and which has in general been considered identical with Willan's Porrigo decalvans). Forhandl. Skand. Natuforsk. fjerde Möde i Christiania den 11—18 Juli 1844, pp. 306—316.

Malmsten, (P.H.) (1845a) Ondskorfvens vegetabiliska natur. (The vegetable nature of Favus. Lecture and case demonstration). Svenska Läkare-Sällskapets sammankomst, 11.3.1845. Förhandl. Svenska Läkare-Sällsk. sammankomster från och med Octob. 1844 till och med Sept. 1845, pp. 58—59, and Hygiea, **7, 11,** 687, 1845.

Malmsten, P. H. (1845b) Trichophyton tonsurans. Hårskärande Mögel. (Trichophyton tonsurans. Hair-cutting mould). Hygiea, **7, 6,** 325—333, 8, 483—487, 1845.

Malmsten, P. H. (1845c) Trichophyton tonsurans, hårskärande mögel. Bidrag till utredande af de sjukdomar som vålla hårets affall. (Trichophyton tonsurans, hair-cutting mould. Contribution to the investigation of the diseases which cause loss of hair). L. J. Hjerta, Stockholm, 1845. Pp. 13.

Malmsten, P. H. (1848) Trichophyton tonsurans, der haarscheerende Schimmel. Ein Beitrag zur Auseinandersetzung der Krankheiten, welche das Abfallen des Haares bewirken. Arch. Anat. Physiol. wiss. Med. herausg. v. Dr. Johannes Müller, **1848,** Jg., 1—19, 1848.

Manfred, Jean (1926) Ett fall av lungaktinomykos. (A case of lung Actinomycosis). Hygiea, **88,** 5, 193—209, 1926.

Marcussen, Poul V. (1937a) Relationship of the Urticarial to the Inflammatory Reaction to Trichophytin. Arch. Derm. Syphilol. **36,** 3, 494—514, 1937.

Marcussen, Poul V. (1937b) Die Bedeutung der Sofortreaktion bei der intracutanen Trichophytinprobe auf der Grundlage von etwa 2000 Untersuchungen. Verh. dtsch. Derm. Ges. Achtzehnte Tagung. Gehalten zu Stuttgart, 18—21 September 1937. Arch. Dermatol. Syphilis, **177,** 87—90, 1937.

Marcussen, Poul V. (1940a) Nyere Undersøgelser over Trichophytin-Reaktion. (Further investigations into the Trichophytin-reaction. Same as Acta dermato-venereologica, **22,** 489, 1941). Dansk Derm. Selsk. Forhandl. Sept. 1939-Maj 1940. Møde, 1.5.1940. Nordisk Med. **9,** 1941: 1, 11, Hospitalstidende, 840—841, 1941.

Marcussen, Poul V. (1940b) Neuere Untersuchungen über Trichophytin-Reaktion. Verh. Dän. Derm. Ges. 1940. 314 Versammlung, 1.5.1940. Acta derm.-vener. **22,** 5, 489—493, 1941.

(Medical Society in Kristiania) Det Medicinske Selskab i Kristiania. (1888) Constitutio epidemica. Forhand. Norske Med. Selsk. i Kristiania i Aaret 1888, pp. 105—106. Norsk Mag. Laegevidensk. **49,** 4 Raekke, **3,** Bilaga, 1888.

Modén, Carl (1935) Om de s.k. aktinomycesliknande kropparna vid kronisk tonsillit. (On the so-called Actinomyces-like granules in chronic tonsillitis). Festskrift utgiven av Svenska Tandläkare-Sällskapet med anledning av dess 75-årsjubileum, 21 november 1935, pp. 71—82. A. B. Fahlcrantz' Boktryckeri, Stockholm, 1935.

Modén, Carl. (1938) Fortsatt undersökning över de s.k.aktinomycesliknande kropparna vid kronisk tonsillit med särskild hänsyn till deras innehåll av trådformiga gramfasta förgrenade mikrober. With English Summary. Further studies in the actinomyces-like granules by chronic tonsillitis with special regard to their content of gram-positive, thread-like, branched microbes. Svensk Tandläkare Tidskr. **31,** 3, 190—203, 1938.

Möller (1902) Fall av favus capillitii totalis. (Case of Favus capillitii totalis). Dermatologiska Sällskapets i Stockholm förhandlingar år 1902. Sammankomst, 30.1.1902. Hygiea, Följd 2, **4,** 467, 1904.

Møller, Henning (1938) Et Tilfaelde af Diabetes mellitus med Gaercellinfektion. (A case of Diabetes mellitus with an infection by yeast cells). Ugeskrift for Laeger, **100,** 9, 223—224, 1938.

Møllgaard, Holger (1931) Über den fraglichen Entwicklungscyclus des Tuberkelbacillus. Beitr. Klin. Tuberk. **77,** 83—120, 1931.

Møllgaard, Holger (1932) Mikrochemische Untersuchungen zur Frage der nicht säurefesten Tuberkelbacillen. Beitr. Klin. Tuberk. **79,** 515—554, 1931/1932.

Moene, Ivar (1939) Behandling av aktinomykose med M & B 693. (Treatment of Actinomycosis with M & B 693. To be publ. in Nord. Medicin). Forhandlinger i det Medicinske Selskab i Bergen 1939. Møte, 9.11.1939. Nordisk Med. **6,** 1940: 2, 14, Medicinsk Rev. 753, 1940.

Moene, Ivar (1940) Behandling av aktinomykose med M & B 693. (Treatment of Actinomycosis with M & B 693). Nordisk Med. **5,** 1940: 1, 8, Medicinsk Rev. 335—337, 1940.

Mossige, K(jell) (1937a) Trichophytie bei Menschen durch Infektion von Silberfüchsen hervorgerufen. (Publiziert in Acta derm.-vener. **19,**, Fase. 3, 1938). Verh. Norw. Derm. Ges. Sitzung, 21.10.1937. Acta derm.-vener. **20,** 3, 327, 1939.

Mossige, Kjell (1937b) See Gjessing, H. Chr. & Mossige, Kjell, 1937.

Mossige, Kjell (1938a) Report on the first observations of Epidermophyton rubrum (Castellani-Bang) in Norway, with an historical review. Acta derm-vener. **19,** 2, 209—219, 1938.

Mossige, Kjell (1938b) See Danbolt, N. & Mossige, Kjell, 1938c.

Mossige, Kjell (1939) See Danbolt, N. & Mossige, Kjell. 1939a.

Mossige, Kjell (1940) Epidermophytia pedis - (”fotsopp”) - dens allergiske symtomer og behandling. (Epidermophytia pedis - Foot Mycosis - its allergic symptoms and treatment. Lecture). Tidsskr. Norske Laegeforening, **60,** 3, 113—128, 1940.

Motzfeldt & Thjøtta (1924) En sjelden lunge-sopinfektion. (A rare fungus infection of the lung). Forhandl. Med. Selsk. i Kristiania 1924, pp. 32—35. Møte, 12.3.1924. Norsk Mag. Laegevidensk. **85,** 5 Raekke, **22,** Bilaga, 1924.

Müller, J. (1842) Über parasitische Bildungen. Arch. Anat. Physiol. wiss. Med. herausg. v. Dr. Johannes Müller, **1842,** Jg., 193—212, 1842.

Müllern-Aspegren, U. (1921) Urétrite et cystite à saccharomycose avec ”pneumaturie”. Acta derm-vener. **2,** 1, 126, 1921.

Näslund, C(arl) and Dernby, K. G. (1923) Untersuchungen über einige physiologische Eigenschaften des Strahlenpilzes. Biochem. Z. **138,** 497—504, 1923.

Naeslund, Carl (1925a) Studies of Actinomyces from the Oral Cavity. Acta path. microbiol. Scand. **2,** 2, 110—140, 1925.

Naeslund, Carl (1925b) Studien über Speichelsteinbildung. Acta path. microbiol. Scand. **2,** 3, 244—275, 1925.

Naeslund, Carl (1926a) Studies of Tartar Formation. Acta path. microbiol. Scand. **3,** 3, 637—677, 1926.

Naeslund, Carl (1926b) Undersökning rörande tandstenens orsaker och uppkomst. (Investigation concerning the causes and formation of tartar). Odontol. Tidskr. **34,** 3, 145—191, 1926.

Naeslund, Carl (1926c) Comparative Study of the Formation of Concretions in the Oral Cavity and in the Salivary Gland and Ducts. Dental Cosmos, **68,** 12, 1137—1146, 1926.

Naeslund, Carl (1927) Über Kalkausfällung durch Mikroorganismen. Biochem. Z. **184,** 1—9, 1927.

NAESLUND, Carl (1928) Tandstenen och dess betydelse för munhålans sjukdomar. (Tartar, and its significance for the diseases of the oral cavity). Nordisk Hyg. Tidskr. **9**,299—306, 1928.

NAESLUND, Carl (1929a) Experimentelle Übertragung der Aktinomykose vom Menschen auf Versuchstiere mittels Reinkulturen der Actinomycesform Wolff-Israel (Act. Israeli). Acta path. microbiol. Scand. **6**, 66—77, 1929.

NAESLUND, Carl (1929b) Fortgesetzte Untersuchungen über die Speichelsteinbildung unter besonderer Berücksichtigung ihres Verhältnisses zur entstehung primärer Speicheldrüsenaktinomykose. Acta path. microbiol. Scand. **6**, 78—96, 1929.

NAESLUND, Carl (1931) Experimentelle Studien über die Ätiologie und Pathogenese der Aktinomykose. Acta path. microbiol. Scand. Suppl. 6, 1931. Pp. 156.

NAESLUND, Carl (1935) Strålsvampsjukdomarnas indelning samt bakteriologiska och serologiska diagnostik. (The classification, as well as the bacteriological and serological diagnostics of Actinomycoses). Nordisk Med. Tidskr. **9**, 12, 440—444, 1935.

NAESLUND, Carl (1939) On What Grounds Can a Classification of Actinomyces and Allied Organisms be Made? Third Int. Congr. Microbiol. New York, September 2—9, 1939. Report of Proceedings, pp. 189—190.

NICOLAYSEN (1893) Aktinomykose, behandlet med Jodkalium. (Actinomycosis treated with Potassium iodide. Case). Forhandl. Med. Selsk. i Kristiania 1893, pp. 83 –84. Møde, 8.3.1893. Norsk Mag. Laegevidensk. **54**, 4 Raekke, **8**, Bilaga, 1983.

NICOLAYSEN, Knud (1918) To kasus av abdominal-aktinomycose. (Two cases of abdominal Actinomycosis). Forhandl. i Kristiania Kirurg. Forening 1918, pp. 149 –151. Møte, 16.9.1918. Norsk Mag. Laegevidensk. **80**, 5 Raekke, 17, Bilaga, 1919.

NICOLAYSEN, N. Aars (1920) Et tilfaelde av kjaeve aktinomykose helbredet med incisioner og store doser K. J. (A case of jaw Actinomycosis healed by incisions and large doses of Potassium iodide). Det Medicinske Selskap i Bergen. Møte, 12.2.1920. Medicinsk Rev. **37**, 5—7, 213—214, 1920.

NICOLAYSEN, N. Aars & SLOLTENBERG, L. (1923) Actinomycosis pulmonum. (Case). Det Medicinske Selskap i Bergen. Møte, 20.12.1923. Medicinsk Rev. **41**, 1, 39—41, 1924.

NICOLL (1871) Tilfaelde af otomycosis. (Case of Otomycosis). Forhandlinger i Bergens Laegeforening i 1871. Møde, 27.10.1871. Norsk Mag. Laegevidensk. 3 Raekke, **2**, 265—266, 1872.

NIELSEN, Jens (1939) Røntgenbehandling af cervico-facial Aktinomykose. (On X-ray treatment of cervico-facial Actinomycosis. Publ. in extenso in Acta Radiologica, **23**, 2, 303—314, 1942). Dansk Radiologisk Selskabs Forhandlinger. 165 Møde, 18.5.1939. Nordisk Med. **4**, 1939: 4, 52, Hospitalstidende, 3841—3842, 1939.

NIELSEN, Ludv(ig) (1894) Pityriasis versicolor hos et 9 Maanader gammelt Barn. (Pityriasis versicolor in a child of 9 months. Communication). Hospitalstidende, **37**, 4 Raekke, **2**, 34, 846, 1894.

NIELSEN, Ludvig (1914) Epilation ved Trichophyti med Ichthyol-Vat-Kalot. (Epilation in Trichophytosis with an Ichthyol-cotton-cap). Dansk Dermatologisk Selskab. 104 Møde, 4.11.1914. Hospitalstidende, **58**, 5 Raekke, **8**, 16, 391—393, 1915.

NIELSEN, Nils (1929) See CHRISTIANSEN, M. & NIELSEN, Nils, 1929.

NORDENSTRÖM, H. (1872) Trenne fall av favus, botade under behandling med karbolsyra. (Three cases of Favus healed by treatment with Phenolic acid). Hygiea, **34**, 3, 143—144, 1872.

NORDENTOFT, Jacob (1914a) Tilfaelde af Aktinomykosis. Meningitis. (Case of Actinomycosis. Meningitis). Jydsk Med. Selskab. 1 Møde, 22.2.1914. Ugeskr. for Laeger, **76**, 19, 903—904, 1914.

NORDENTOFT, Jakob (1914b) Kasuistisk Meddelelse om nogle Tilfaelde af Aktinomykose med Omtale af Forsøg paa Röntgenbehandling. (Casuistical communications on some cases of Actinomycosis with mention of an attempt at X-ray treatment). Jydsk Medicinsk Selskab. 1 Møde, 22.2.1914. Nordisk Tidsskr. Terapi, **12**, 272,—276 1913—14.

NORDENTOFT, Jakob (1933) Aktinomycose i Thisted Amt. (Actinomycosis in the Thisted District). Jydsk Med. Selsk. Forhandl. 1932—1933, pp. 158—164. 107 Møde, 2.4.1933. Hospitalstidende, **76,** Bilaga, 1933.

NORGAARD, A. (1920a) Om Vaekst af Penicillier i Morfinopløsninger. (On the growth of Penicilli in Morphine solutions. Abstract. Publ. in extenso in Ugeskr. for Laeger, **82,** 1235, 1920). Dansk Selskab for Intern Medicin. 32 Møde, 26.3.1920. Ugeskr. for Laeger, **82,** 40 1275, 1920.

NORGAARD, A. (1920b) Om Vaekst af Skimmelsvampe i Morfinopløsning. Meddelelse paa 32. Møde i Selskab for intern Medicin. (On the growth of mould fungi in Morphine solution. Communication). Ugeskr. for Laeger, **82,** 39, 1235—1236, 1920.

NORLÉN, Sune (1925) Zur Kenntnis der Aktinomykose. Zwei Fälle von Fremdkörper im Mundboden. Svenska Läkaresällskapets Handlingar, Acta Soc. Med. Suecanae, **51,** 4, 227—237, 1925.

NORRLIN, C. L. Lennart (1915) Bidrag till kännedomen om och behandlingen af perforerande enkla ventrikel- och duodenalsår. (Contriburion to the knowledge and the treatment of perforated simple ulcers of the ventricle and the duodenum). Thesis, Stockholm, 1915. Almqvist & Wiksells Boktryckeri, A.B. Uppasla 1915. Pp. 276.

NYSTRÖM, A. (1890) Skäggsvamp-sjukan eller Sycosis parasitaria. (Mycosis of the beard or Sycosis parasitaria. Case demonstration, and lecture). Förhandl. Svensk. Läkaresällsk. sammankomster år 1890, pp. 203—207. Sammankomst, 21.10.1890. Hygiea, 52, Bilaga, 1890.

ODENIUS (1883) Om actinomyces iakttagen hos en kalkun. (On Actinomycosis observed in a turkey). Protokoll vid Fysiografiska Sällskapets (i Lund) sammanträde, 14.3.1883.

ODENIUS, Quoted by LJUNGGREN, C. A., 1895.

ØRSKOV, J. (1920) En Jagttagelse angaaende Slaegtskaber mellem Streptothricherne og Bakterierne. (An observation concerning the relationship between Streptothrices and bacteria). Hospitalstidende, **63,** 48, 753—763, 1920.

ØRSKOV, J. (1922a) Methode til Rendyrkning af Bakterier fra een Celle samt Fremgangsmaade til direkte Forfølgelse af Bakterievaekst paa fast Substrat. (Method for the isolation of Bacteria in pure culture from single cells and procedure for the direct tracing of bacterial growth on a solid medium. Same as J. Bact. **7,** 537, 1922). Hospitalstidende, **65,** 6, 86—98, 1922.

ØRSKOV, J. (1922b) Method for the Isolation of Bacteria in Pure Culture from Single Cells and Procedure for the Direct Tracing of Bacterial Growth on a Solid Medium. J. Bact. **7,** 6, 537—549, 1922.

ØRSKOV, J. (1923) Investigations into the Morphology of Ray Fungi. Thesis, Copenhagen, 1923. Levin and Munksgaard, Copenhagen, 1923. Pp. 171.

ØRSKOV, J. (1938) Untersuchungen über Strahlenpilze, reingezüchtet aus dänischen Erdproben Zbl. Bakt. Parasitenk. 2 Abt., **98,** 16/20, 344—357, 1938.

ØRSKOV, J. (1939) On the Morphology of Some Species of Micromyces. Third Int. Congr. Microbiol., New York, September 2—9, 1939. Report of Proceedings, p. 179.

ORLA-JENSEN, S. (1909) Die Hauptlinien des natürlichen Bakteriensystems. Cbl. Bakt. Parasitenk. 2 Abt. **22,** 11—13, 305—346, 1908—1909.

OTTANDER, Olle (1933a) 2. Fall av Oidiomykos. (2. Case of Oidiomycosis). See BERGMAN, R., DANIELSON, C. G., OTTANDER, Olle & SCHNELL, Å., 1933b.

OTTANDER, Olle (1933b) Ett fall av Oidiomykos. Diskussionsinlägg till DANIELSON, C. G.: Fall av pneumonomykos. (A case of Oidiomycosis. Contribution to a discussion. Pp. 403—407). Sammankomst, 16.5.1933. Svenska Läkaresällsk. Förhandl. år 1933, pp. 389—414.

PALM, Björn & HINRICSSON, Harry (1940) En avtrycksmetod för studiet av mikroorganismer på olika ytor. Zusammen fassung: Eine Abdrucksmethode zum Studium der Mikroorganismen auf gewissen Substraten. Tidskr. Militär Hälsovård, **65,** 4, 147—151, 1940.

PANUM, P. L. (1876) Studier over Gäring och Forrådnelse med särligt Hensyn til de mikroskopiske Organismers Andel i Fermentvirkningerne. (Studies on fermentation and putrification with special regard to the role of the microscopical organisms in the fermentative activity). Nordiskt Med. Ark. **8,** 25, 1—35, 1876.

Petersen, Hj. (1914) 2 Tilf. af Pityriasis versicolor. (Two cases of Pityriasis versicolor). Dansk Dermatologisk Selskab. 104 Møde, 4.11.1914. Hospitalstidende, **58,** 5 Raekke, 8, 16, 397—398, 1915.

Plesner, Marie (1936, 1939, 1940) See Jersild, O. & Plesner, Marie, 1936, 1939, 1940.

Pontoppidan, B. (1915) Et Tilfaelde af Favus corporis. (A case of Favus corporis). Hospitalstidende, **58,** 5 Raekke, 8, 29, 729—731, 1915.

Pontoppidan, B. (1919) 2 Tilfaelde af Kerion celsi med likenoide Trikofytider. (Two cases of Kerion celsi with lichenoid Trichophytids). Dansk Dermatologisk Selskab. 135 Møde, 5.2.1919. Hospitalstidende, **62,** 12, 378—380, 1919.

Pontoppidan, B. (1920) Trichophytia manus. (Case). Dansk Derm. Selsk. Forhandl. i Oktober 1919-Maj 1920, p. 36. 145 Møde, 4.2.1920. Hospitalstidende, **63,** Bilaga, 1920.

Pontoppidan, Erik (1885a) Beretning fra Kommunehospitals Poliklinik for Hudsygdomme og veneriske Sygdomme for Treaaret d. 1ste Marts 1882 til d. 1ste Marts 1885. (Report from the Outpatient Department for Skin and Venereal Diseases of the Municipal Hospital for the three-year-period from the 1st of March, 1882 to the 1st of March, 1885. Pp. 297—301). Hospitals-Tidende, 3 Raekke, **3,** 247—309, 1885.

Pontoppidan, Erik (1885b) Hudsygdommene og de veneriske Sygdommene. En kortfattet Fremstilling. (Skin and venereal diseases. A short presentation. Dermatomycoses, pp. 76—82). P. G. Philipsens Forlag, Kjøbenhavn, 1885. Pp. 167.

Pontoppidan, Erik (1890) Hudsygdommene og de veneriske Sygdomme. En kortfattet Fremstilling. Anden omarbeidede Udgave. (Skin and venereal diseases. A short presentation. Second revised edition. Dermatomycoses, pp. 87—92). P. G. Philipsens Forlag, Kjøbenhavn, 1890. Pp. 194.

Pontoppidan, Erik (1891) Therapeutiske Meddelelser. Ti tusende Tilfaelde af Hydsygdomme og veneriske Sygdomme, behandlede 1885—90. (Therapeutic notices. Ten thousand cases of skin and venereal diseases treated from 1885 to 1890. Dermatomycoses, pp. 21—23). P. G. Philipsens Forlag, Kjøbenhavn, 1891. Pp. 110.

Pontoppidan, Erik (1896) Hudsygdommene og de veneriske Sygdommene. En kortfattet Fremstilling. Tredje gjennemsete Udgave. (Skin and venereal diseases. Third revised edition. Dermatomycoses, pp. 82—87). Det Nordiska Förlag, Kjøbenhavn, 1896. Pp. 191.

Prip, Holger (1894) To Tilfaelde af animal Trikofyti hos Mennesket. (Two cases of animal Trichophytosis in man). Hospitalstidende, **37,** 4 Raekke, **2,** 31, 765—772, 1894.

Quensel, Ulrik (1929) Multipla plasmacellsinfiltrat i tunntarmen. Egenartad mykotisk (?) sjukdomsform. Deutsches Referat: Multiple Plasmazelleninfiltrate im Dünndarm. Eine eigenartige mykoticshe (?) Krankheitsform. Finska Läkaresällsk. Handl. **71,** 9, 661—671, 1929.

Ramm (1892) Tilfaelde av Aktinomykose paa Kindet. (Case of Actinomycosis on the cheek, with demonstration of preparations by Kjerschow). Forhandl. Norske Med. Selsk. i Kristiania 1892, p. 222. Møde, 2.11.1892. Norsk Mag. Laegevidensk. **53,** 4 Raekke, **7,** Bilaga, 1892.

Rasch, C. (1898) Meddelelser fra Kommunehospitals Poliklinik for Hudsygdomme og veneriske Sygdomme. Dec. 1893-Dec. 1897. I. Hudsygdomme. 1. Actinomycosis cutis. 25. Trichophytia. (Communications from the Outpatient Department for Skin and Venereal Diseases at the Municipal Hospital. Dec 1893-Dec. 1897. I. Skin diseases. 1. Actinomycosis cutis, pp. 713—717. 25. Trichophytosis, pp. 1142—1146). Hospitalstidende, **41, 4** Raekke, **6,** 28, 713—722, 42, 1133—1146, 1898.

Rasch, C. (1905) Hudens Sygdomme og deres Behandling. Hudsygdomme fremkaldte af Svampe. (Diseases of the skin and their treatment. Skin diseases caused by fungi, pp. 44—75). Gyldendalske Boghandel, Nordisk Forlag København, Kristiania, 1905. Pp. 545.

Rasch, C. (1907a) Alopeci og Mb. mentalis (pyromani) efter Röntgenbehandling af Mikrospori i Haarbunden. (Alopecia and Morbus mentalis, Pyromania, following X-ray treatment of Microsporia in the ciapillitium). Dansk Dermatologisk Selskab. 53 Møde, 1.5.1907. Hospitalstidende, **50,** 4 Raekke, **15,** 33, 869—870, 1907.

Rasch, C. (1907b) Röntgenbehandling af Trikofyti. (X-ray treatment of Trichophytosis). Hospitalstidende, **50,** 4 Raekke, **15,** 48, 1299—1300, 1907.

Rasch, C. (1908) Röntgenbehandling af Trikofyti. (X-ray treatment of Trichophytosis). Hospitalstidende, **51,** 5 Raekke, **1,** 3, 71, 1908.

Rasch, C. (1909) Hudens Sygdomme og deres Behandling. Anden Udgave. Hudsygdomme fremkaldte af Svampe. (Diseases of the skin and their treatment. Second edition. Skin diseases causes by fungi, pp. 41—70). Gyldendalske Boghandel, Nordisk Forlag, København 1909. Pp. 501.

Rasch, C. (1913a) Erythematøs Favus i Ansigtet hos et nyfødt Barn. Smitte fra modern. (Erythematous Favus of the face in a newborn child. Contamination from the mother. Case). Dansk Dermatologisk Selskab. 95 Møde, 7.5.1913. Hospitalstidende, **56,** 5 Raekke, **7,** 4, 127, 1914.

Rasch, C. (1913b) Tilfaelde af saakaldet Blastomykose paa Haandryggen. (Case of so-called Blastomycosis on the back of the hand). Dansk Dermatologisk Selskab. 98 Møde, 3.12.1913. Hospitalstidende, **56,** 22, 730—731, 1914.

Rasch, C. & Kissmeyer, A. (1914) Podningsforsøg paa Marsvin. (Attempt at inoculation into Guinea pigs. Communication concerning the case 3.12.1913). Dansk Dermatologisk Selskab. 102 Møde, 13.5.1914. Hospitalstidende, **56,** 44, 1337, 1914.

Rasch, C. (1915) Om sekundaere, likenoide Trikofytider ved Kerion celsii. Lichen spinulosus trichophyticus. (On seconderay lichenoid Trichophytids with Kerion celsi). Dansk Dermatologisk Selskab. 111 Møde, 3.11.1915. Hospitalstidende, **59,** 5 Raekke, **9,** 14, 324—327, 1916.

Rasch, C. (1916a) Secondary Lichenoid Trichophytides in Association with Kerion Celsi, Lichen spinulosus trichophyticus. Brit. J. Derm. **28,** 1, 9—13, 1916.

Rasch, C (1916b) Tilfaelde af saakaldet Blastomykose paa Haandryggen. (Case of so-called Blastomycosis on the back of the hand. Case from 3.12.1913). Dansk Dermatologisk Selskab. 117 Møde, 4.10.1916. Hospitalstidende, **60,** 40, 989, 1917.

Rasch, C. (1916c) Kerion med likenoide Trikofytider. (Kerion with lichenoid Trichophytids). Dansk Dermatologisk Selskab. 119 Møde, December, 1916. Hospitalstidende, **61,** 22, 745—746, 1918.

Rasch, C. (1917a) Kerion med likenoide Trikofytider. (Kerion with lichenoid Trichophytids). Dansk Dermatologisk Selskab. 120 Møde, Febr., 1917. Hospitalstidende, **61,** 23, 778, 1918.

Rasch, C. (1917b) Trikofyti med Kerion, likenoide og skaellende Trikofytider og diffus Pigmentering af Huden. (Trichophytosis with Kerion, lichenoid and squamous Trichophytids and diffuse pigmentation of the skin). Dansk Dermatologisk Selskab. 120 Møde, Febr., 1917. Hospitalstidende,**61,** 23, 778—779, 1918.

Rasch, C. (1918) Tilfaelde af Sporotrichosis. (Case of Sporotrichosis). Dansk Dermatologisk Selskab. 129 Møde, 3.4.1918. Hospitalstidende, **61,** 41, 1406—1407, 1918.

Rasch, C. (1919a) Klinische Bemerkungen über Trichophytia profunda, Kerion Celsi. (Abstract by the Author. Publ. in extenso in Ugeskr. for Laeger, **82,** 443, 120, and Brit. J. Derm. **32,** 317, 1920. Forhandl. Nordisk Derm. Foren. fjerde Møde i København, 10—12 Jvni, 1919, pp. 97—101.

Rasch, C. (1919b) Kliniske Bemerkninger om Trichophytia profunda, Kerion Celsi. (Clinical remarks on Trichophytia profunda, Kerion Celsi. Same in English in Brit. J. Derm. **32.** 317, 1920). Meddelt paa Nordisk Dermatologisk Forenings 4-de Møde i København i Juni 1919. Ugeskr. for Laeger. **82,** 14, 443—449, 1920.

Rasch, C. (1919c) Clinical Remarks on Trichophytia profunda, Kerion Celsi. Delivered at the Fourth Meeting of the Nordisk Dermatologisk Forening, Copenhagen, June 1919. Brit. J. Derm. **32,** 11, (385), 317—325, 1920.

Rasch, C. (1921) Om de af Achorion gypseum Bodin fremkaldte Hudaffektioner. (On the skin affections caused by Achorion gypseum Bodin). Dansk Derm. Selsk. Forhandl. i Oktober 1920-Maj 1921, pp. 61—63. 157 Møde, 6.4.1921. Hospitalsstidende, **64,** Bilaga, 1921.

Rasch, C. (1922) Eczema marginatum Hebra and other Mycoses resembling Eczema. Proc. Fifth Meeting Northern Derm. Soc. Stockholm, 6—8 June, 1922. Acta derm.-vener. **3,** 3—4, **315—344,** 1922.

Rasch, C. (1923)Favus i Skaegget. (Favus in the beard. Case). Dansk Derm. Selsk. Forhandl. 1922—1923, p. 30. 172 Møde, 7.2.1923. Hospitalstidende, **66,** Bilaga, 1923.

Rasch, C. (1924a) Tilfaelde af Mikrospori, M. Audouini, hos en Voxen samt Tilfaelde af "Bald ringworm" hos et Barn, framkaldt af samme Svamp. Case of Microsporia, M. Audouini, in an adult, and a case of "Bald Ringworm" in a child, caused by the same fungus). Dansk Derm. Selsk. Forhandl. 1923—1924, pp. 45—46. 182 Møde, 7.5.1924. Hospitalstidende, **67,** Bilaga, 1924.

Rasch, C. (1924b) Kliniske Bemerkninger om Trichophyton rosaceum Sabouraud 1909. (Clinical remarks about Trichophyton rosaceum Sabouraud 1909). Festskrift i Anledning af Dansk Dermatologisk Selskabs 25-Aars Bestaaen. Bibliotek for Laeger, **116, 363—369,** 1924.

Rasch, C. (1924c) En Skimmelsvamp, Scopulariopsis brevicaulis var. homis Brumpt & Langeron 1910, som Aarsag til Neglelidelse. (A mould fungus, Scopulariopsis brevicaulis var. hominis Brumpt & Langeron **1910,** as the cause of nail affection. Case. Same in German in Derm. Wschr., **82,** 551, 1926). Dansk Derm. Selsk. Forhandl. 1924—1925, pp. 19—21. 185 Møde, 3.12.1924. Hospitalstidende, **68,** Bilaga, 1925.

Rasch, C. (1924d) Ein Schimmelpilz, Scopulariopsis brevicaulis var. hominis Brumpt und Langeron 1910, als Ursache eines Nagelleidens. Verh. Dän. Derm. Ges. 185 Sitzung, 3.12.1924. Derm. Wschr. **82, 16, 551,** 1926.

Rasch, C. (1925a) Tilfaelde af Trikofyti hos et lille Barn, Trichophyton album Sabouraud 1909. (Case of Trichophytosis in a small child, Trichophyton album Sabouraud 1909). Dansk Derm. Selsk. Forhandl. 1924—1925, pp. 61—62. 189 Møde, 6.5.1925. Hospitalstidende. **68,** Bilaga, 1925.

Rasch, C. (1925b) Tilfaelde af Trikofyti sundet under Behandling med Jodkalium invendig. (Case of Trichophytosis healed by internal treatment with Potassium iodide). Dansk Derm. Selsk. Forhandl. 1924—1925, pp.72—73. 189 Møde, 6.5. 1925. Hospitalstidende, **68,** Bilaga, 1925.

Rasch, C. (1927) Hudens Sygdomme og deres Behandling. Tredie omarbejdede og forøgede Udgave. II. Hudsygdomme fremkaldte af Svampe. (Diseases of the skin and their treatment. Third revised and augmented edition. II. Skin diseases caused by fungi, pp. 54—101). Gyldendalske Boghandel. Nordisk Forlag, København, 1927.

Rasmussen, Anker Frode (1883) Om Dyrkning af Mikroorganismer fra Spyt af sunde Mennesker. (On the cultivation of microorganisms from the sputum of healthy persons). Thesis, Copenhagen, 1883. Jacob Lund, København, 1883. Pp. 136.

Reenstierna, John (1912) Ueber die Kultivierbarkeit des Lepraerregers und die Uebertragung der Lepra auf Affen. Deutsch. Med. Wschr. **38,** 2, 38, 1784—1785, 1912.

Reenstierna, John (1913) Über die Kultivierbarkeit und Morphologie des Lepraerregers und die Übertragung der Lepra auf Affen. Arch. Dermatol. Syphilis, **116,** 3, 480—554, 1913.

Reenstierna, J(ohn) (1923) La bacille de la lépre. 3^e Conférene Internationale de la Lépre, Strasbourg, Juillet, 28—31, 1923. Communications et débats.

Reenstierna. J(ohn) (1926a) Reproduction expérimentale du mycétome chez le lapin. Arch. Inst. Pasteur, Tunis, **15,** 2, 101—107, 1926.

Reenstierna, J(ohn) (1926b) Sur la vie non-acidorésistante du microorganisme de la tuberculose. Arch. Inst. Pasteur, Tunis, **15,** 4, 309—317, 1926.

Reenstierna, John (1927) Experimental Reproduction of Mycetoma in Rabbits. Festskrift tillägnad Professor J. E. Henschen. Acta med. Scand. **65,** 566—572, 1926/27.

Reenstierna, John (1937) A Fourth Orientation on the Therapeutic Value of an Anti-Leprosy Serum. Acta med. Scand. Suppl. 85, 1937. Pp. 106.

Reenstierna, J(ohn) (1938) See Gullberg, Evert, 1938.

Reisz, C. (1896) Trösksvamp, Oidium albicans, på Ventrikelns Slemhinde. (Thrush fungus, Oidium albicans, on the mucous membranes of the ventricle). Nordiskt Med. Ark. **1,** 7, 1—10, 1986.

Renander, Axel (1937) Le Traitement Radiologique de l'Actinomycose. Acta radiol. Suppl. 35, 1937. Pp. 75.

Reyn (1905a) Favus og Folliculitis barbae behandlede med Röntgenstraaler. (Favus and Folliculitis barbae treated with X-rays. Cases). Dansk Dermatologisk Selskab. 40 Møde, 1.3.1905. Hospitalstidende, **48,** 4 Raekke, **13,** 28, 650—651, 1905.

Reyn (1905b) Et Tilfaelde af Trichophytia capitis behandlet med Röntgenlys. (A case of Trichophytia capitis treated with X-rays). Dansk Dermatolologisk Selskab. 43 Møde, 4.10.1905. Hospitalstidende, **49,** 4 Raekke, **14,** 23, 649—650, 1906.

Reyn, Axel (1909) Behandling af Hudsygdomme med Röntgenstraaler. (Treatment of skin diseases with X-rays. Trichophytosis and Favus, pp. 1022—1027). Hospitalstidende, **52,** 5 Raekke, **5,** 33, 1017—1027, 1909.

Riising, Jørgen (1921) Et sjaeldent Tilfaelde af Aktinomykose. (A rare case of Actinomycosis). Ugeskr. for Laeger, **83,** 25, 833—838, 1921.

Rischel, Axel (1931) 2 Tilfaelde af Aktinomycosis. (Two cases of Actinomycosis). Ugeskr. for Laeger, **93,** 18, 484—486, 1931.

Rossander, Carl J. (1891) Actinomycosis. (Case reports, p. 13). Årsrapport från Kongl. Serafimerlasaretter för år 1891.

Rossander, Carl J. (1892) Actinomycosis. (Case reports, pp. 14—15). Årsrapport från Kongl. Serafimerlasarettet för år 1892.

Rostrup, Ove (1909) Nogle Undersøgelser over Luftens Inhold af Svampekim. (Some investigations into the air content of fungous germs. Investigation accomplished during 1903/04). Bot. Tidsskr. **29,** 1, 32—41, 1908—1909.

Rovsing, Thorkild (1887) Et Tilfaelde af Aktinomykose. (A case of Actinomycosis). Hospitals-Tidende, 3 Raekke, **5,** 38, 889—895, 1887.

Rovsing, Thorkild (1889) Om Aktinomykosen hos Mennesket. (On Actinomycosis in man. Review). Bibliotek for Laeger, **81,** 6 Raekke, **19,** 74—107, 1889.

Rovsing, Thorkild (1920) Aktinomykose - Straalesvamp. (Pp 209—215. Chapter in) Lärobok i Kirurgi. I. Allmän Kirurgi. (Textbook of Surgery. I. General Surgery). Edited by Borelius, J., Nicolaysen, J. and Rovsing, T. M.P. Madsens Boghandel, København, 1920.

Runeberg, B. (1914) Kvinna med aktinomykos i vänstra bröstet. (A woman with Actinomycosis of the left mamma). Finska Läkaresällskapet, 7.11.1914. Finska Läkaresällsk. Handl. **56,** 2, 12, 1673, 1914.

Runeberg, (J.W.) (1892) Förevisning av en pat. med bukaktinomykos. (Demonstration of a patient with abdominal Actinomycosis). Finska Läkaresällskapets sammanträde 3.12.1892. Finska Läkaresällsk. Handl. **35,** 1, 61—62, 1893.

Sabouraud (1907) Röntgenbehandling af Trikofyti. (X-ray treatment of Trichophytosis. Letter in French). Hospitalstidende, **50,** 4 Raekke, **15,** 52, 1398—1403, 1907.

Sabouraud (1908) Röntgenbehandling af Trikofyti. (X-ray treatment of Trichophytosis. Letter in French). Hospitalstidende, **51,** 5 Raekke, **1,** 7, 181—183, 1908.

Saeves, Inga (1915) Experimentelle Beiträge zur Dermatomykosenlehre. Arch. Dermatol. Syphilis, **121,** 2, 161—236, 1915.

Schaumann, Jörgen (1917a) Recherches sur le lupus pernio et ses relations avec les sarcoïdes cutanées et sous-cutanées. Nordiskt Med. Ark. **49,** Avd. 2, Ark. Inre Med. Häft 6, 17, 1—81, 1917.

Schaumann, J(örgen) (1917b) Etudes bactériologiques sur le lupus pernio et les sarcoïdes cutanées. Kungl. Boktryckeriet A. Norstedt & Söner, Stockholm, 1917. Pp. 39.

Schaumann Jörgen (1930) Discussion. Comptes rendus des séances, p. 548. 8^e Congr. Int. Derm. Syphiligr. Copenhague, 5—6 Août, 1930.

Schaumann, J(örgen) and Introzzi, P. (1931) Sulla eziologia e sulle alterazioni sistematiche degli organi ematopoietici del lupus eritematose acuto. Haematologica, 1 Archivio, **12,** 7, 635—717, 1931.

Schaumann, Jörgen (1934) Observations cliniques bacteriologiques et sérologiques pour servir á l'étiologie de la lymphogranulomatose bénigne. Réunion Dermatologique de Strasbourg. Séance spéciale du 13 Mai 1934 consacrée á l'étude des sarcoïdes. Bull. Soc. Franç. Dermatol. Syphiligr. **41,** 1296—1322, 1934.

Schaumann, Jörgen (1936) Lymphogranulomatosis Benigna in the Light of Prolonged Clinical Observations and Autopsy Findings. Brit. J. Derm. **48,** August-September, 399—446, 1936.

Scheen (1912) Actinomycosis faciei (Case). Forhandl. Med. Selsk. i Kristiani 1912, pp. 98—99. Møte, 24.4.1912.Norsk Mag. Laegevidensk. **73,** 5 Raekke, **10,** Bilaga, 1912.

Schlasberg (1929) Two cases of trichophytosis cutis. Transactions of the Society of Dermatologists in Stockholm for the year 1929. Meeting, 11.9.1929. Acta derm.-vener., **11,** 4 332, 1930.

Schmiegelow, E. (1896) Bidrag till Patogenesen af den saakaldte sorte Tunge. (Contribution to the pathogenesis of the so-called Black tongue). Ugeskr. for Laeger, 5 Raekke, **3,** 23, 528—543, 1896.

Schnell, Åke (1933a) Fall av mukopurulent bronkitis förorsakat av en Oidium-art. (Case of mucopurulent Bronchitis caused by an Oidium species). Tuberkuloseföreningens förhandlingar, 11.5.1933. Hygiea, **96.** 18, 649—651, 1934.

Schnell, Åke (1933b) See Bergman, R., Danielson, C. G., Ottander, O. and Schnell, Åke, 1933b.

Schou, Jens (1895) Et Tilfaelde af Underlivsaktinomykose helbredt ved Jodkalium. (A case of Actinomycosis in the hypogastrium healed with Potassium iodide). Ugeskr. for Laeger, 5 Raekke, **2,** 50, 1174—1178, 1895.

Schultén, af (1877) Aspergillus flavescens i yttre hörselgången. (Aspergillus flavescens in the external auditory canal. Case). Möte, 24.11.1877. Finska Läkaresällsk. Handl. **20,** 4, 317—318, 1878.

Schultén, af (1893) Ett fall av bukaktinomykos. (A case of abdominal Actinomycosis). Finska Läkaresällskapets sammanträde, 4.3.1893. Finska Läkaresällsk. Handl. **35,** 296—297, 1893.

Schultén, af (1897) Förevisning av en pat. opererad för temporomaxillar aktinomykos. (Demonstration of a patient, operated on for a temporomaxillary Actinomycosis). Finska Läkaresällskapets sammanträde, 24.4.1897. Finska Läkaresällsk. Handl. **39:** 1,6.782, 1897.

Sederholm (1893) Favus af ovanligt vidsträckt utbredning. (Favus of an unusually wide extension). Förhandl. Svenska Läkare-Sällsk. sammankomster år 1893, ppl 27—29. Sammankomst, 31.1.1893. Hygiea, **55,** Bilaga, 1893.

Sederholm (1903) Behandlingen af herpes tonsurans capitis. (Treatment of Herpes tonsurans capitis). Derm. Sällsk, i Stockholm förhandl. i år 1903. Sammankomst, 29.10.1903. Hygiea, Följd 2, **4,** 11, 1290, 1904.

Selander, N. E. (1887) Luftundersökningar vid Vaxholms fästning okt. 1885—juli 1886. (Air examination at Vaxholm's fortress from Oct. 1885 to July 1886). Thesis, Stockholm, 1887. Kongl. Bocktryckeriet P. A. Norstedt & Söner, Stockholm. 1887, and Bihang till Kongl. Svenska Vetenskaps-Akademiens Handlingar, **13,** Afdelning 2, Nr. 9, 1888. Pp. 38.

Sell, A. (1879) Tilfaelde af "Lingua nigra". (Case of "Lingua nigra"). Hospitals-Tidende, 2, Raekke, **6,** 49, 877—980, 1879.

Sell, A. (1885) Tilfaelde af "Lingua nigra". (Case of "Lingua nigra"). Hospitals-Tidende, 3 Raekke, **3,** 4, 87—89, 1885.

Serck-Hanssen, Trygve & Swensson, N.V. (1927) Aktinomykose. (Actinomycosis). Medicinsk Rev. **44,** 2, 71—86, 1927.

Sjöbring, (1901) Om archiplasmats struktur i svulstceller och om dess förhållande till de intracellulära jästsvamplika parasitformerna. (On the structure of archiplasma with tumour cells, and on its relations to the intracellular yeat fungus-like parasite forms). Läkaresällskapets i Lund förhandlingar. Sammanträde, 24.9.1901. Hygiea, Följd 2,**2,** 3, 341—348, 1902.

Sjögren, T. (1904) Om användning af Röntgenstårlar vid behandling af herpes tonsurans capillitii. (On the application of X-rays in the treatment of herpes tonsurans capillitii). Allmänna Svenska Läkartidn. **1,** 44, 775—778, 1904.

Slomann, H. C. (1893) To Tilfaelde af Kerion Celsi. (Two cases of Kerion Celsi). Hospitalstidende, **36,** 4 Raekke, **1,** 33, 845—850, 1893.

Söderbaum, P. (1893) Några kirurgiska fall från Falu lasarett. Fall av Aktinomykosis. (Some surgical cases from the Hospital at Falun. Case of Actinomycosis, pp. 227—230). Upsala Läkarefören. Förhandl. **28,** 4, 215—232, 1892—93.

Söderlund, Gustaf (1909) Om primär spottkörtelaktinomykos. Ett bidrag till frågan om ansikts- och halsaktinomykosens ingångsport och etiologi (On primary Actinomycosis of the salivary gland. A contriburion to the question of the port of entrance and the aetiology of the Actinomycosis of the face and neck). Upsala Läkarefören. Förhandl. Ny följd. **14,** 3—4, 266—295, 1908—1909.

Söderlund, Gustaf (1913a) Über die primäre Aktinomykose der Speicheldrüsen. Nordiskt Med. Ark. **46,** 3:e följden, 13, Afd. 1, Kirurgi, Nr. 13, 1—186, 1913.

Söderlund, Gustaf (1913b) Über die primäre und isolierte Aktinomykose der Speicheldrüsen. Dtsch. med. Wschr. **39:** 2, 34, 1632—1634, 1913.

Söderlund, Gustaf (1915) Über die primäre Aktinomykose der Speicheldrüsen. (Abstract by the author in Nord. Med. Ark., 1913). Upsala Läkarefören. Förhandl. Ny följd, **20,** 5, 318—330, 1914—1915.

Söderlund, Gustaf (1919) En undersökning öfver den s.k. spottstenssjukdomen (''Sialolithiasis'') speciellt dess etiologi. (An investigation into the so-called disease of concretion in the salivary gland, Sialolithiasis, particularly its aetiology). Förhandl. Nordisk Kirurg. Fören. 12:e Möte i Kristiania, 3—5 Juli, 1919, pp. 74—87.

Söderlund, Gustaf (1921) Einige neue Beiträge zur Kasuistik der primären Speicheldrüsenaktinomykose. Acta chirurg. Scand. **53,** 3, 189—229, 1920/21.

Söderlund, Gustaf (1928) Die Speichelsteinkrankheit (''Sialolithiasis'') und ihr Verhalten zu der primären und duktogenen Speicheldrüsen-Actinomykose. Acta chirurg. Scand. **63,** Suppl. 9, 1928. Pp. 237.

Söderlund, Gustav (1932) Aktinomykosen - Strålsvampsjukdomen. (Actinomycosis, pp. 229—235. Chapter in)Nordisk Lärobok i Kirurgi. I. Allmän Kirurgi. Andra omarbetade upplaga. (Northern Textbook of Surgery. I. General Surgery. Second revised edition) Edited by Ingebrigtsen, R., Petrén, G. and Schaldemose, V. Levin and Munksgaards Forlag, København, 1932.

Söderlund, Gustav (1940) Aktinomykosen - Strålsvampsjukdomen. (Actinomycosis, pp. 250—257. Chapter in) Nordisk Laerebok i Kirugi. I. Almindelig Kirurgi. Tredje omarbeidede Utgave. (Northern Textbook of Surgery. I. General Surgery. Third revised edition). Edited by Ingebrigsten, R., Kjaergaard, S. and Petrén, G. Ejnar Munksgaard, København, 1940.

Söndegaard, Karl (1935) Tre Tilfaelde af Lungeaktinomykose. (Three cases of lung Actinomycosis). Dansk Radiologisk Selskab Sept. 1935. Hospitalstidende, **80,** 27, 801—810, 1937.

Sopp (Johan-Olsen), Olav (1913) Straalesoppen fra Toten. Straalesoppens opholdssted og mugfrugt. (Actinomyces from Toten. The occurrence and the fructification organs of the Actinomyces). Norsk Mag. Laegevidensk. **74,** 5 Raekke, **11,** 1025—1047, 1913.

Sopp (Johan-Olsen), Olav (1926) Nogen bemerkninger til dr. med. Gundersen's artikel: ''Sopsporecylindre i urin''. (Some remarks to Dr. med. Gundersen's paper). Norsk Mag. Laegevidensk. **87, 3,** 247—249, 1926.

Stecksén, Anna (1900) Studier öfver Curtis' blastomycet från svulst-etiologisk synpunkt. (Studies on Curtis' Blastomyces from the tumour-etiological point of view). Thesis, Stockholm, 1900. Kungl. Boktryckeriet. P. A. Norstedt & Söner, Stockholm, 1900. Pp. 88.

Stoltenberg, L. (1923) See Nicolaysen, N. A. & Stoltenberg, L. 1923.

Strandberg, (James) (1913) Fall von Onychomycosis favosa. Verh. zweiten Kongr. Nordischen Derm. Ver. zu Stockholm, 5—7 Juni, 1913. Nordiskt Med. Ark. **46,** 3:e Följden, **13,** Afd. 2, Inre Medicin, Bilaga, 1913.

Strandberg, (James) (1914) Ett fall av herpes tonsurans å händer och fötter. (A case of Herpes tonsurans of the hands and feet). Derm. Sällsk. Stockholm förhandl. år 1914. Sammanträde 4.11.1914. Hygiea, **77,** 2, 55—56, 1915.

Strandberg, James (1917) Om en svampsjukdom å händer och fötter. (On a fungus disease of the hands and feet). Festskrift till Professor Dr. Alfred Pettersson. Svenska Läkaresällsk. Handl. **43,** 3, 913—916, 1917.

Strandberg, (James) (1920) A case of trichophyton, resembling microsporon.

Proc. Derm. Soc. in Stockholm. Meeting, 13.10.1920. Acta derm.-vener. **1,** 3—4 474—475, 1920.

Strandberg, (James) (1931) Trichophytia profunda capitis + Lichen trichophyticus corporis. (Case). Verh. Derm. Ges. zu Stockholm. Sitzung, 11.4.1931. Acta derm.-vener. **12,** 4, 378—379, 1931.

Sundewall, Fredr. (1871) Fall af Trichophyton behandlad med campher. (Case of Trichophytosis treated with Camphor). Upsala Läkareförenings sammanträde, 3.11.1871. Upsala Läkarefören. Förhandl. **7,** 2, 173—175, 1871—1872.

Sundberg, Carl (1897) Mikroorganismerna från läkarens synpunkt. Läro- och kort handbok. Actinomyces. Hypho- och blastomykoser. (The microorganisms from the physician's point of view. Textbook and a short manual. Actinomyces, pp. 487—492. Hypho- and Blastomycoses, pp. 563—568). W. Schultz, Upsala, 1897. Pp. 589.

Sundberg, Henri (1919) Ett fall av primärt? aktinomykom i omentum majus. (A case of primary? Actinomycoma of the omentum majus). Svenska Läkaresällsk. Kirurg. sektionens förhandl. 1919 —1920. Sammanträde, 19.9.1919. Hygiea **82,** 12, 412—413, 470—473, 1920.

Svitzer, E. (1845) Praktiske Jagttagelser. 11. En sielden Sygdom i Haaret. (Practical observations. A rare disease of the hair, pp. 44—54). Bibliotek for Laeger, **42,** Ny Raekke, **12,** 1—54, 1845.

Swensson, N.V. (1927) See Serck-Hanssen, T. & Swensson, N.V., 1927.

Sydow, v., Gert (1936) Fall av lungaktinomykos. (Case of lung Actinomycosis). Svenska Fören. Med. Radiologi Förhandl. Sammanträde, 16.5.1936. Nordisk Med. Tidskr. **14,** 2, 49, 2003—2004, 1937.

Sylvest, Bent (1949) The Incidence of Dermatophytes in Denmark. Acta derm. —.vener. **29,** 3, 225—232, 1949.

Takala, M. (1922) Aktinomykosista ja sen hoidista. (On Actinomycosis and its treatment). Duodecim, 38, 6—7,261—264, 1922.

Tawast-Rancken, Saima (1938) Jod-iontoforesbehandling vid aktinomykos. (Jodine iontophoresis treatment of Actinomycosis). Kirurgföreningens i Finland förhandl. Årsmöte, 18.11.1938. Nordisk Med. **1,** 1939: 1, 12, Nordisk Med. Tidskr. 884, 1939.

Tawast-Rancken, Saima (1939a) Aktinomykoosin jodi-iontoforeesihoidosta. Deutsches Referat: Über die Jod-Jontophoresebehandlung der Aktinomykose. Duodecim, **55,** 8, 622—628, 1939.

Tawast-Rancken, Saima (1939b) Om jod-iontoforesbehandling av aktinomykos. (On Jodine iontophoresis treatment of Actinomycosis. Swedish translation of the paper from Duodecim, **55,** 622, 1939). Nordisk Med. **3,** 1939: 3, 38, Duodecim, 2954—2956, 1939.

Thesen (1909) Tilfaelde af aktinomykose. (Case of Actinomycosis). Forhandl. Med. Selsk. i Kristiania 1909, pp. 234—235. Møde, 24. 11.1909. Norsk Mag. Laegevidensk. **70,** 5 Raekke, **7,** Bilaga, 1909.

Thjøtta, (Th.) (1924a) See Motzfeldt & Thjøtta, 1924.

Thjøtta, Th. and Gundersen, Edv. (1924b) En streptothrix-art isolert fra blodet hos en rheumatiker. (A Streptothrix species isolated from the blood of a rheumatic patient). Festskrift till Professor Med. Hed. Dr. Klas Sondén. Nordisk Hyg. Tidskr. **5,** 5—10, 249—262, 1924.

Thjøtta, T(h.) and Gundersen, E(dv.) (1925) A streptothrix isolated from the blood in a case of acute rheumatism with remarks upon the classification of ray fungi. J. Bact. **10,** 1, 1—12, 1925.

Thjøtta, Th. (1939) See Bøe, J., Hartmann, O. & Thjøtta, Th., 1939.

Thomsen, Axel (1937) Occupational diseases in Danish veterinary surgeons. A Survey. Infectious Diseases. 1. Ringworm, pp. 731—733. Skand. vet. Tidskr. **27,** 728—755, 1937.

Trier, F. (Editor) (1867) Nogle nyere Bidrag til Kundskaben om Svampe, der frembringe Sygdomme hos Mennesket, (Some further contributions to the knowledge of fungi which cause diseases in man. A survey). Ugeskr. for Laeger, 3 Raekke, **4,** 5, 57—76, 1867.

Troilius, S.G. (1863) Om Favus, Herpes tonsurans och Alopecia areata samt deras företeelser inom Sverige. Svar på af Kongl. Sundhets-kollegium begärda upplysningar. (On Favus, Herpes tonsurans, and Alopecia areata, and their manifestations in Sweden. Reply to the information requested by the Royal Board of Health). Hygiea, **25,** 10, 401—418, 1863.

Tscherning (1893) Et Tilfaelde af Perityphlitis actinomycotica. (A case of Perityphlitis actinomycotica). Med. Selsk Forhandl. Møde, 7.3.1893. Hospitalstidende, **36,** 4 Raekke, **1,** 18, 520—521, 1893.

Turesson, Oscar (1916) Fall av actinomykos. (Case of Actinomycosis). Tuberkuloseläkareföreningens förhandl. Sammanträdet, 19.1.1916. Hygiea, **79,** 10. 491—495, 1917.

Vedeler (1907a) Blastomyceter i blodet. (Blastomycetes in blood. Demonstration of microscopical slides). Forhandl. Med. Selsk. i Kristiania 1907, p. 152. Møde, 25.9.1907. Norsk Mag. Laegevidensk. **68,** 5 Raekke, **5,** Bilaga, 1907.

Vedeler (1907b) Blastomyceter i blodet. (Blastomycetes in blood. Lecture. To be publ. in Norsk Mag. Laegevidensk.) Forhandl. Med. Selsk. i Kristiania 1907, p. 195. Møde, 23.10.1907. Norsk Mag. Laegevidensk. **68,** 5 Raekke, **5,** Bilaga, 1907.

Vedeler (1907c) Blastomyceter i blodet. (Blastomycetes in blood. Read before the Medicinsk Selskab i Kristiania, 23.10.1907). Norsk Mag. Laegevidensk. **69,** 5 Raekke, **6,** 968—976, 1908.

V(ennerhol) M. J(ohn) (1929) Yrkesinfektioner hos veterinären. Favus och Trichophyti. (Occupational infections in veterinary surgeons. Favus and Trichophytosis, p. 246). Svensk vet. Tidskr. **34,** 8—9, 235—253, 1929.

Videbach, P. (1914) Et Tilfaelde af Aktinomykose begyndene som akut Appendicit. (A case of Actinomycosis starting as acute Appendicitis). Jydsk Medicinsk Selskab. 1 Møde, 22.2.1914. Ugeskr. for Laeger, **76,** 19, 900—903, 1914.

Vosbein, Erich B. (1939) Mykotisk Fingeraffektion med sekundaert "Erythema multiforme". (Mycotic affections of the finger with secondary "Erythema multiforme"). Dansk Derm. Selsk. Forhandl. 302 Møde, 1.2.1939. Nordisk Med. **6,** 1940: 2,14, Hospitalstidende, 683, 1940.

Voss, J. A. (1923a) Et tilfaelde av general blastomykose, meningit. (A case of general Blastomycosis. Meningitis. To be publ. in Norsk Mag. Laegevidensk.). Forhandl. Med. Selsk. i Kristiania 1923, p. 20. Møte, 21. 2. 1923. Norsk Mag. Laegevidensk. **84,** 5 Raekke, **20,** Bilaga, 1923.

Voss, J.A. (1923b) Et tilfaelde av general blastomykose, forløpande under billedet av en meningit. (A case of general Blastomycosis, running its course under the picture of a Meningitis. Demonstrated in the Medicinsk Selskab i Kristiania, 21.2.1923). Norsk Mag. Laegevidensk. **84,** 5 Raekke, **20,** 6, 550—560, 1923,

Voss, J.A. (1934) Et tilfelle av sporotrichosis pulmonum. Résumé français: Un cas de sporotrichose des poumons. Meddelt i den Norske Patologforening 1934. Norsk Mag. Laegevidensk. **96,** 1,14—16, 1935.

Wahlberg (1852) Om en i Nerike nu gängse endemisk sjukdom uppkommen genom den osunda beskaffenheten af förra årets rågskörd. See Levin, P.A., 1852a, and Hök, 1852. (Report concerning the rye sample sent in by LEVIN). Hygiea, **14,** 2, 83—86, 1852.

Waldenström, (J.A.) (1869) Dermatomycosis. Eczema marginatum, pp. 87—97. Onychomycosis, pp. 97—104. (Cases). Upsala Läkareförenings sammanträde, 15.10.1869. Upsala Läkarefören. Förhandl. **5,** 2, 87—104, 1869—1870.

Waldenström. J.A. (1872) Polikliniken i Upsala stad under läsåret 1870—1871. 4. Onychomycosis, Svampnagel. (The policlinic at Upsala during the acade mical year of 1870—1871. Onychomycosis, pp. 179—183). Upsala Läkfören. sammanträde, 25.10.1872. Upsala Läkarefören. Förhandl. **8,** 2, 176—183, 1872—1873.

Welander, Edvard (1900) Einige Versuche, Herpes tonsurans capillitii, Trichophytie, mit Wärme zu behandlen. Festschrift gewidmet Moriz Kaposi. Arch. Dermatol. Syphilis, **1900,** Ergänzungsband, 35—58, 1900.

Wendt, von, Dora (1926) Beitrag zur Kenntnis der Bakteriologie der Ulcusperitonitiden. Eine klinisch-bakteriologische Studie. Arb. Path. Inst. Univ. Helsingfors (Finland). Neue Folge, **4,** 1/2, 131—210, 1926.

Westling, Rich. (1911) Über die grünen Spezies der Gattung Penicillium. Ark. Bot. **11,** 1, 1—156, 1911.

Widröe, Sofus (1912a) Et tilfaelde av sporotrichose. (A case of Sporotrichosis.

To be publ. in Nord. Med. Ark. 1912). Forhandl. Med. Selsk. i Kristiania 1912, pp. 30—34. Møte, 28.2.1912. Norsk Mag. Laegevidensk. **73,** 5 Raekke, **10,** Bilaga, 1912.

Widröe, Sofus (1912b) Ein Fall von Sporotrichosis. Nordiskt Med. Ark. **45,** 3:e Följden, **12,** Afd. 1, Kirurgi, Häft 3, 6, 1—4, 1912.

Winge, E. (1871) Onychomycosis. (Case). Forhandl. Norske Med. Selsk. i Kristiania i 1871, pp. 128—133. Møde, 24.5.1871. Norsk Mag. Laegevidensk. 3 Raekke, **1,** Bilaga, 1871.

Wissing, O. (1928) Tilfaelde af Actinomycose i Lungen (Pleura?) Behandlet med Røntgenstraaler. (Case of lung Actinomycosis - pleura? - treated with X-rays). Dansk Radiol. Selsk. Forhandl. 1928, pp. 51—52. 64 Møde, 12.12.1928. Hospitalstidende, **72,** Bilaga, 1929.

Åkerman, Jules (1891) Actinomycosis hominis. Hygiea, **53,** 5—6, 595—607, 1891.

Addendum

Here are added some publications that had unfortunately been left out in the main review.

In 1910 and 1914 af Heurlin (F) had observed Saccharomycetes to occur also in the genital canal of non pregnant women.

Harbitz (N) summed up his experiences regarding the pathological anatomy of Actinomycosis in lectures that were issued in print in 1923. His material had consisted of a total of 133 Norwegian cases, 2/3 of which had occurred in men and 1/3 in women. Of the different clinical types Harbitz (N) had seen cervico facialis in 51%, thoraco-pumonalis in 20%, abdominalis in 26%, and primary cutaneous Actinomycosis in 3%. More reports on Actinomycosis were met with from Denmark by Videbach (D), 1940, and from Sweden by Behring (S), 1933, Ahltorp (S), 1934, and Sundberg (S), 1934. According to Sundberg(S) in the course of 1933 57 men and 29 women had in Sweden been admitted for clinical ward for Actinomycosis.

Bibliography

Ahltorp, Gideon (1934) Ein Fall von Aktinomykose in den Beckenorganen einer Frau. Acta obstet. et gynec. Scand. **13**, 1, 81—92, 1933/34.

Behring, Ivar (1933) The Occurrence of Actinomycosis of the Stomach and the Duodenum. Liber Gratulatorius in Honorem John Forssman, pp. 18—30. Acta path. et microbiol. Scand. **10**, Suppl. 16, 1933.

Harbitz, Francis(1923) Forelaesninger i patologisk anatomi over syfilis, lepra og aktinomykose. (Lectures into the pathological anatomy of Syphilis, Leprosy, and Actinomycosis). Steenske Forlag, Kristiania 1923. Pp.71.

Heurlin, af, Manu (1910) Bakteriologische Untersuchungen des Keimgehaltes im Genitalkanale der fiebernden Wöchnerinnen mit Berücksichtigung der Gesamtmorbidität im Laufe eines Jahres. Thesis, Helsingfors 1910. Kommisionsverlag der Akademischen Buchhandlung, Helsingfors 1910. Pp.618.

Heurlin, af, Manu (1914) Bakteriologische Untersuchungen der Genitalsekrete der nichtschwangeren und nichtpuerperalen Frau vom Kindes- bis ins Greisenalter unter physiologischen und gynäkologisch-pathologischen Verhältnissen. Verlag von S. Karger, Berlin 1914. Pp.226.

Sundberg, Stig (1934) Aktinomykos i munhålan jämte beskrivning av ett fall. (Actinomycosis of the oral cavity, with the description of a case). Svenska Tandläk. Tidskr. **27**, 3, 381—395, 1934.

VIDEBECH, H. (1940) A Case of Larynx Actinomycosis. (Publ. in Nordisk Med, **9**, 571—1941) Transactions of the Danish Oto-Laryngological Society 1939—40. 294 Meeting, 13.1.1940. Acta Oto-Laryngologica, **30**, 2, 176, 1942.

VIDEBECH, H. (1940) Tilfaelde af Larynxaktinomykose. (A case of larynx Actinomycosis). Dansk Oto-laryngologisk Selskabs Forhandlinger. Møde, 13.1.1940. Nordisk Med. **9**, 8, Hospitalstidende, 771—572, 1941.

GROWTH AND NUTRITION OF A STRAIN OF GYMNOASCUS REESSII IN SHAKEN FLASKS

by

H. H. KUEHN & P. F. CROSBY

Grain Processing Corporation, Muscatine, Iowa, U.S.A.

(14.XI.1959)

In an endeavor to develop sounder bases for taxonomy of the Gymnoascaceae, KUEHN (1955) initiated his studies of the developmental morphology of these fungi. Subsequent investigations revealed that certain characteristics of developmental morphology could be utilized to place taxonomy within the family on a sound basis. However, within recent years in instances involving two different species, *Pseudoarachniotus roseus* and *Gymnoascus reessii*, the possibility was suggested that combined application of structural and developmental morphological characteristics might not always suffice to identify species with certainty. One example, involving *P. roseus*, will be treated in a separate publication. The other example, that of *G. reessii*, will be discussed at this time.

During the last several years the senior author has had occasion to examine numerous strains of *Gymnoascus reessii*, all of which possessed similar structural characteristics with regard to ascospores, ascocarps, etc. Moreover, the features of the developmental processes were identical among these strains. However, while most of the strains possessed similar cultural characteristics, two strains were distinctly different in this regard. These two strains were those designated as RSA 60 and RSA 495. Strain RSA 60 was described in detail earlier by KUEHN (1956) and the reader is referred to that article for the complete description. Suffice it to state here that cultures of this strain on YpSs agar are characterized by a green colonial coloration, whereas the usual strains of *G. reessii* are typified by hyaline to white colonies prior to ascocarp formation. Strain 495 was received from Dr. R. K. BENJAMIN with the following notation, "received from MMe J. NICOT, Mus. Nat., Paris, her 180A.40. She did not give full collection data. I received the culture from her in Jan., 1957". Colonies of strain RSA 495 are characterized by a prominent yellow color which colors the entire colony prior to ascocarp formation. In both strains 60 and 495, the ascocarp normally has a less extensively branched peridium and presents the appearance illustrated by KUEHN (1956). Other strains of *G. reessii*, however, typically form ascocarps which have quite extensively branched peridia and more nearly resemble the illustration of BENJAMIN (1956).

The purpose of the present investigation was twofold. One purpose was to initiate studies of the nutrition of various strains of *Gymnoascus reessii* in an endeavor to determine whether such physiological data might be utilized for taxonomic purposes to compare strains of slight morphological or cultural variation and to thus determine their specific affinities. It is our intention to compare other strains of *G. reessii* in a comparable investigation in order to determine whether within a given species all strains can be classified into the same nutritional type or whether subgenera might exist based upon nutritional differences.

A second purpose was to develop a defined, optimum, synthetic medium for growth of RSA 495. This basal medium would then be employed as a basal medium in subsequent investigations regarding the nature of specific metabolites formed in submerged cultures of this strain.

MATERIALS AND METHODS

The culture of *Gymnoascus reessii* used throughout this study was the strain RSA (Rancho Santa Ana Botanic Garden) 495 already discussed. The experimental procedures were similar to those presented and discussed in detail by KUEHN & CROSBY (1959) for *Pseudoarachniotus roseus*. Briefly, the procedures were as follows. Cultures were grown in duplicate using 100 ml medium in 500 ml Erlenmeyer flasks which were incubated usually for 2½ to 3½ days on a reciprocating shaker. Preliminary experiments showed that the fungus failed to grow at 37°C, therefore, all further experiments were incubated at 28°C. The experimental flasks were inoculated with 2% of 3—10 day old submerged growth brought up in inoculum flasks of YpSs medium. Following the desired period of incubation the mycelium was removed from the culture broth by centrifugation and the dry weight was used as an index of growth. The pH of the supernatant was recorded as the terminal pH.

Since YpSs and Sabouraud's media were known to support good growth of *Gymnoascus reessii* and other Gymnoascaceae, these media were employed in preliminary studies to determine the optimum initial pH and rate of growth as a function of time. The composition of the media was as follows: YpSs — 1.5% soluble starch, 0.4% yeast extract, 0.1% K_2HPO_4 and 0.05% $MgSO_4$ 7 H_2O; Sabouraud's —4% maltose, 1% peptone. Further experiments were performed to establish the assimilability of various nitrogen and carbon sources, requirements and optimum levels of various vitamins, purines and pyrimidines as well as $MgSO_4$ 7 H_2O and K_2HPO_4. Most of the earlier experiments were carried out using modifications of the YpSs medium. The basal medium in each experiment was modified to conform with results of previous experiments with the result that a synthetic medium was developed which was optimum for growth of *G. reessii*. The exact composition of the medium used in each experiment will be presented with the discussion of that experiment in the appropriate position in the text.

RESULTS AND DISCUSSION

Preliminary experiments were conducted to determine the effect of the pH range from 3.0—9.0 on growth of this ascomycete. Using the YpSs medium in an 86 hour incubation period approximately equivalent growth was obtained with all pH levels higher than 3.0, as shown in Table I. Comparison of the terminal pH levels would indicate that the fungus brought about an alteration in pH of the medium during growth. In spite of this modification of the pH the favorable pH still ranged from 5.0 to 8.0. It was evident that the initial pH within the range 5—8 was not critical for growth and therefore an initial pH of 7.0 was adopted for all further experiments.

TABLE I.

Influence of initial pH upon growth

Initial pH	Terminal pH	Mycelium weight mg/100 ml
3.0	4.1	282
4.0	5.1	776
5.0	6.2	771
6.0	6.8	794
7.0	7.5	782
8.0	7.6	828
9.0	8.0	793

To determine the rate of growth as a function of time in each of two media 12 replicate flasks of YpSs and 12 flasks of Sabouraud's medium were placed on the shaker simultaneously. Duplicate flasks were removed from the shaker at approximately daily intervals. The results (Figure 1) indicated that the maximum growth was attained in 3 days with YpSs and in 5 days with Sabouraud's medium and that thereafter the weight of mycelium decreased with the onset of autolysis. Based upon this information further experiments were terminated following 2½ to 3½ days of incubation.

In order to study the nitrogen assimilation of *Gymnoascus reessii* various nitrogen-containing compounds were substituted at a concentration of 0.4% for yeast extract in the YpSs medium and the flasks were incubated for 81 hours. Table II presents the results with the compound listed in descending order of utilization. Potassium

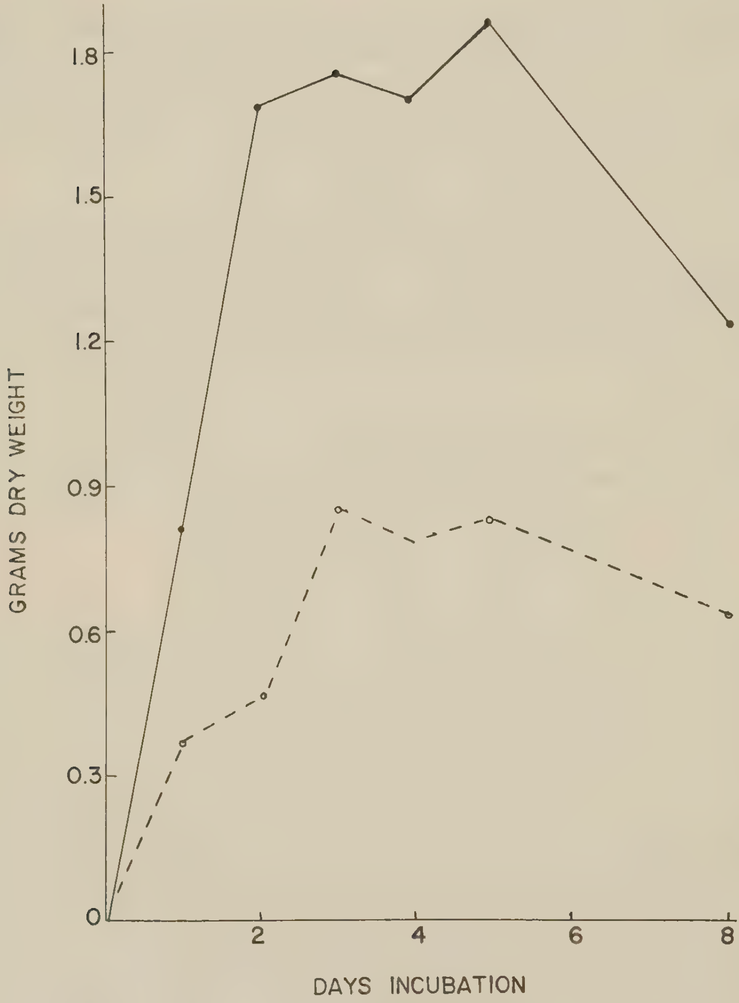


Fig. 1. Rate of growth in each of two media; solid line — Sabouraud's medium, broken lines — YpSs medium.

nitrate was the best defined source of nitrogen followed by *dl*-alpha alanine. The terminal pH levels with ammonium acetate, ammonium nitrate and urea indicate that in this unbuffered medium a pH effect might be responsible for poor growth. Table III contains results of an 83 hour experiment in which certain of the better nitrogen sources were supplied at higher concentrations. A third experiment regarding nitrogen sources was performed in which multiple combinations of defined nitrogen sources were tested. In none of these three types of experiments did any defined nitrogen compound,

TABLE II.

Utilization of various nitrogen- containing compounds.

Compound	Terminal pH	Mycelial weight mg/100 ml
peptone	7.5	851
yeast extract	7.2	825
KNO_3	8.2	661
dl-alpha alanine	7.4	645
l-proline	7.8	604
corn steep liquor	7.7	602
$NaNO_3$	8.1	592
glycine	8.1	566
dl-asparagine	8.0	552
dl-tryptophan	5.8	542
$(NH_4)_2HPO_4$	6.0	518
l-tyrosine	7.0	510
beta-alanine	8.1	495
dl-valine	7.3	478
phenyl alanine	6.8	473
$Ca\ (NO_3)_2$	7.6	471
l-arginine	7.7	435
dl-aspartic acid	7.7	418
dl-methionine	5.0	405
ammonium tartrate	4.5	346
l-glutamic acid	5.9	343
l-cystine	4.2	334
l-cysteine	4.1	314
dl-isoleucine	6.0	311
ammonium acetate	8.6	294
lysine	5.5	261
dl-threonine	6.7	247
NH_4NO_3	4.0	241
controls	7.1	179
urea	9.2	107

TABLE III.

Influence of increased concentrations and combinations of certain nitrogen-containing compounds upon growth.

compound	Concentration	Terminal pH	Mycelial weight mg/100 ml
yeast extract	0.8%	7.2	1009
KNO_3 plus *dl*-alpha alanine	0.4% each	7.6	775
KNO_3	0.8%	7.9	731
dl-asparagine	0.8%	8.1	673
KNO_3 plus *dl*-alpha alanine	0.2% each	7.4	644
glycine	0.8%	7.8	563
$(NH_4)_2HPO_4$	0.8%	6.6	478

used singly or in combination, induce the growth response elicited by yeast extract or peptone.

The basal medium for the studies regarding carbon assimilation consisted of 1.5% carbon source, 0.4% KNO3, 0.4% *dl*-alpha alanine, 0.1% K_2HPO_4 and 0.05% $MgSO_4.7H_2O$. In Table IV the compounds are listed in descending order of utilization. Although dextrin supported the most growth during this 72 hour experiment, maltose was selected as the carbon source in further experiments since the dextrin was not definitely known to be of a high degree of purity. It was surprising that sucrose should be decidedly less available than glucose, fructose or maltose.

TABLE IV.

Assimilability of various carbon-containing compounds.

Compound	Terminal pH	Mycelial weight mg/100 ml
dextrin	7.2	909
soluble starch	7.6	862
maltose	7.4	850
glucose	7.5	660
fructose	7.5	601
xylose	7.0	541
mannose	7.4	518
tartaric acid	8.5	471
lactose	8.2	470
sorbitol	8.2	450
sodium acetate	8.9	415
sucrose	8.4	352
citric acid	8.4	260
calcium gluconate	7.9	206
controls	8.6	170
sorboses	6.6	154

Since yeast extract as well as peptone elicited much more growth than any defined nitrogen source it was quite evident that some growth factor was limiting. A survey of various vitamins, purines and pyrimidines was made to establish the requirement of these compounds for growth of *G. reessii* in a basal medium of 4% maltose, 0.5% KNO_3 and 0.5% *dl*-alpha alanine. The results after 96 hours incubation (Table V) indicated that each of the compounds tested stimulated growth of the fungus, and that the degree of stimulation often was dependent upon the concentration.

Although further investigations were indicated regarding growth factor requirements, it was decided to establish first the exact optimum concentrations of maltose, KNO_3, $MgSO_4.7H_2O$ and K_2HPO_4 in a basal medium containing those growth factors which seemed to be most stimulatory for growth.

TABLE V.

Influence of certain growth factors upon growth.

Compound	Concentration mg per cent	Terminal pH	Mycelial weight mg/108 ml
thiamin	0.2	6.1	358
,,	0.4	6.1	357
biotin	0.2	5.6	454
,,	0.4	6.3	525
calcium pantothenate	0.2	5.8	443
riboflavin	0.2	5.9	457
pyridoxine	0.2	6.0	549
adenine	1.0	5.8	571
,,	4.0	6.1	452
xanthine	1.0	5.7	479
,,	4.0	5.7	689
guanine	1.0	5.6	540
,,	4.0	6.0	535
uracil	1.0	6.0	474
,,	4.0	6.4	362
controls	—	5.9	311

TABLE VI.

Influence of various concentrations of $MgSO_4 . 7H_2O$ *and* K_2HPO_4 *upon growth.*

		per cent $MgSO_4 . 7H_2O$				
		0.0	0.1	0.2	0.3	0.4
per cent K_2HPO_4	0.0	737*	955	790	969	963
	0.1	832	1398	1707	1686	1518
	0.2	792	1354	1824	1525	1491
	0.3	824	1496	1699	1599	1830
	0.4	938	1561	1652	1781	1571

* denotes mg mycelium per 100 ml medium.

Table VI illustrates the results of an experiment designed to establish the requirements for additional $MgSO_4.7H_2O$ and K_2HPO_4. The medium contained 4% maltose, 0.5% KNO_3 0.5% *dl*-alpha alanine, 0.4 mg% biotin, 0.2 mg% pyridoxine, 1 mg% adenine, 1 mg% guanine and 4 mg% xanthine. The experiment was terminated following a 60 hour incubation period. It was quite apparent that in this medium supplemented with growth factors growth was more than two times as great as in the YpSs medium and about equivalent to the growth in Sabouraud's medium following a longer period of incubation (see Figure 1). Perusal of the data in Table VI would indicate that the addition of at least 0.2% of each of K_2HPO_4 and $MgSO_4.7H_2O$ is required for maximum growth. Lack of addition of either or both of these inorganic compounds seriously retarded production of mycelium.

It was felt that in a medium containing various growth factors additional inorganic nitrogen (as KNO_3) might suffice in lieu of *dl*-alpha alanine. Therefore in an experiment to determine optimum levels of KNO_3 and maltose, the basal medium was modified as follows: 0.2% each of K_2HPO_4 and $MgSO_4.7H_2O$, 1 mg% each of adenine and guanine, 4 mg% xanthine, 0.4 mg% biotin and 0.2 mg% pyridoxine. Table VII presents the data following a 60 hour period of incubation. It was apparent in this medium supplemented with growth factors that with proper amounts of maltose and KNO_3 the amount of growth equalled that obtained when *dl*-alpha alanine was used in conjunction with KNO_3 (see Table VI). Therefore in further experiments KNO_3 was used as the sole source of nitrogen. The data illustrate that 5% maltose and 2% KNO_3 are optimum for growth.

TABLE VII.

Influence of various concentrations of KNO_3 and maltose upon growth.

		per cent maltose					
		0.0	1.0	2.0	3.0	4.0	5.0
per cent KNO_3	0.0	32*	222	264	537	604	590
	1.0	98	756	918	1089	1396	1536
	2.0	117	1002	1018	1460	1387	1831
	3.0	144	1068	1077	1456	1657	1707
	4.0	174	1057	1347	1386	1684	1348

*denotes mg mycelium per 100 ml medium.

Three experiments were undertaken to determine the optimum concentrations or combinations of those vitamins, purines and pyrimidines which had been demonstrated to be most stimulatory to growth (see Table V). In each of the experiments the basal medium contained 4% maltose, 2% KNO_3, 0.2% $MgSO_4.7HO_2$, 0.2% K_2HPO_4, plus certain of the growth factors not to be varied plus various amounts of the compounds to be tested. All three experiments were incubated for 64 hours. In one experiment all possible combinations of 1 mg% uracil, 1 mg% adenine, 1 mg% guanine and 4 mg% xanthine were tested using the single and/or multiple omission technique. The basal medium also contained 0.4 mg% biotin and 0.2 mg% pyridoxine. Results showed that when none of these 4 compounds were present growth amounted to 1,136 mg/100 ml medium, while when all 4 were present 1,359 mg/100 ml medium were produced. All other combinations resulted in amounts of growth which were intermediate between these two levels.

According to results shown in Table V a level of 4 mg% adenine inhibited growth compared to the 1 mg% concentration. The same was true for uracil and guanine. However, in the case of xanthine 4 mg% was more stimulatory than 1 mg%. It was not known how much greater the concentration of xanthine should be to permit optimum growth, and, therefore, an experiment was performed to establish this concentration. The basal medium also contained 1 mg% each of adenine, uracil and guanine, 0.4 mg% biotin and 0.2 mg% pyridoxine. It was evident (Table VIII) that 5 mg% xanthine was the optimum concentration.

TABLE VIII.

Influence of xanthine concentration upon growth

Concentration mg per cent	Terminal pH	Mycelial weight mg/100 ml
0	7.6	1148
1.0	7.6	1016
2.0	7.5	1005
3.0	7.6	1089
4.0	7.5	1005
5.0	7.5	1494
6.0	7.5	850
7.0	7.5	783
8.0	7.5	657
9.0	7.4	625

Another experiment was designed to determine optimum levels of biotin and pyridoxine. The basal medium also contained 1 mg% each of adenine uracil and guanine and 5 mg% xanthine. The data showed (Table IX) that 0.8 mg% biotin and 0.2 mg% pyridoxine were sufficient to give optimum yields of mycelium.

TABLE IX.

Influence of concentration of biotin and pyridoxine upon growth.

mg per cent pyridoxine	mg per cent biotin	Terminal pH	Mycelial weight mg/100 ml
—	0.2	7.4	1097
—	0.4	7.7	1137
—	0.6	7.7	1516
—	0.8	7.7	1669
—	1.0	7.6	1042
0.2	—	7.7	1251
0.4	—	7.5	1163
0.6	—	7.6	1262
0.8	—	7.6	1252
1.0	—	7.5	1075

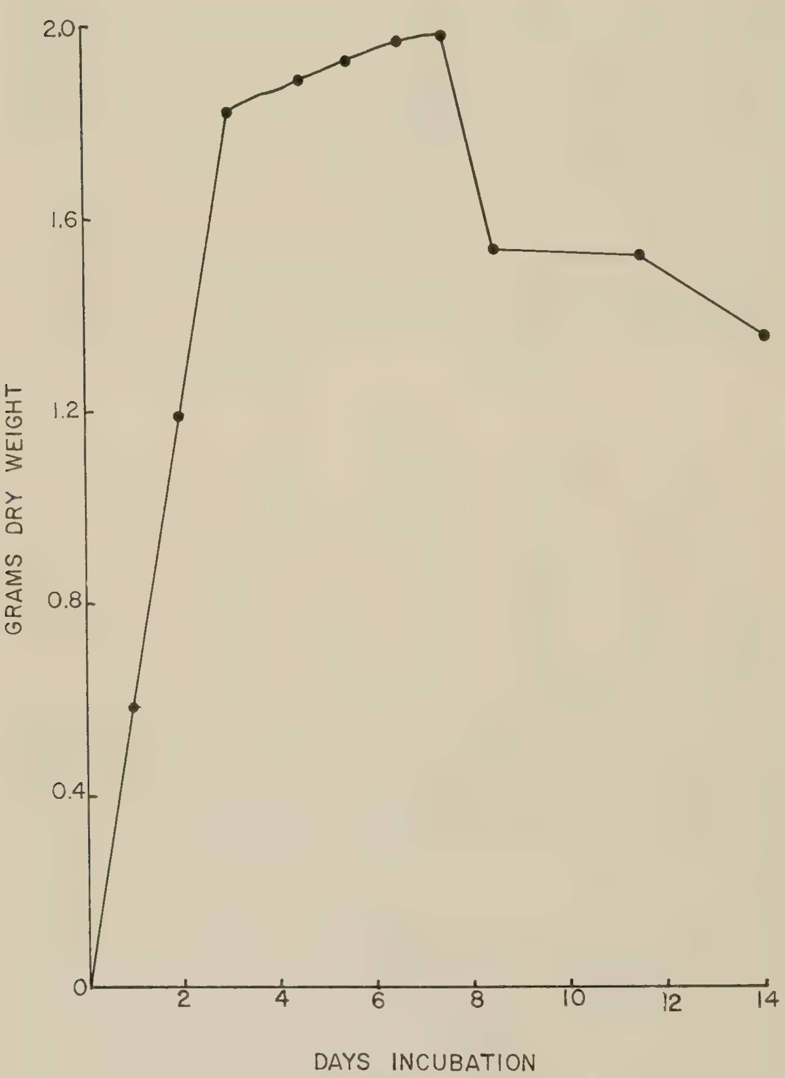


Fig. 2. Rate of growth in the optimum synthetic medium.

Experimentation had now resulted in a medium which could at least tentatively be designated as an optimum growth medium. This medium had the following composition: 5% maltose, 2% KNO_3 0.2% K_2HPO_4, 0.2% MgSO4.$7H_2O$, 1 mg% each of adenine, uracil and guanine, 5 mg% xanthine, 0.8 mg% biotin and 0.2 mg% pyrido xine. Not only was the growth rate in this medium (Figure 2) more rapid than in Sabouraud's and YpSs media (see Figure 1) but the maximum weight of mycelium attained was greater than in either of the „natural" media.

SUMMARY

The fundamental nutritional requirements of one strain of *Gymnoascus reessii* were established. Growth was measured on a dry weight basis. The optimum medium for growth consisted of 5% maltose, 2% KNO_3, 0.2% K_2HPO_4, 0.2% $MgSO_4.7H_2O$, 1 mg% adenine, 1 mg% uracil, 1 mg% guanine, 5 mg% xanthine, 0.8 mg% biotin and 0.2 mg% pyridoxine. An initial Ph range of 5.0—8.0 was found to permit optimum growth.

ZUSAMMENFASSUNG

Die Grunderfordernisse der Ernährung eines Stammes von *Gymnoascus reessii* sind festgestellt worden. Das Wachstum war an der trockenen Gewichtsgrundlage gemessen. Der optimale Nährboden für das Wachstum bestand in 5% Maltose, 2% KNO_3, 0.2% K_2HPO_4, 0.2% $MgSO_4$. $7HO_2$, 1 mg% Adenine, 1 mg% Uracil, 1 mg% Guanine, 5 mg% Xanthine, 0.8 mg% Biotin und 0.2 mg% Pyridoxine. Eine Anfangsbreite der Ph von 5.0—8.0 erlaubte ein optimales Wachstum.

LITERATURE CITED

BENJAMIN, R. K. 1956. A new genus of the Gymnoascaceae with a review of the other genera. Aliso **3**: 301—328.

KUEHN, H. H. 1955. Observations on Gymnoascaceae. I. *Myxotrichum uncinatum* and a new species of *Myxotrichum*. Mycologia **47**: 533—545.

KUEHN, H. H. 1956: Observations on Gymnoascaceae. III. Developmental morphology of *Gymnoascus reessii*, a new species of *Gymnoascus* and *Eidamella deflexa*. Mycologia **48**: 805—820.

KUEHN, H. H., & P. F. CROSBY. 1959. Growth and nutrition of *Pseudoarachniotus roseus* in submerged culture. Mycopath. et Mycol. Appl. **11**: 109—127.

LIBRI NOVI

SCHWARTZ, L., TULIPAN, L. & BIRMINGHAM, D. J.: Occupational Diseases of the Skin. Third edition, thoroughly revised, 189 illustrations and 2 plates in color, 981 pp. Lea & Febiger, Philadelphia, Pa., 1957.

A philosopher who once reviewed one of his friends' work said that all was well and outstanding up to the chapter which covered his own field of interest. This sentence may already reverberate what will soon follow.

The first edition of this standard work, written by the senior author and Dr. TULIPAN, was published in 1939. It filled a long-felt gap, because there was no text in the American literature embracing the field of occupational diseases of the skin. With the ever progressing development of the chemical industry and technology, particularly with the outbreak of the Second World War, the specific knowledge of occupational hazards of the skin, their prevention and treatment became of paramount importance. With the proper insight that one man alone was hardly able to master the vast amount of facts and figures in the field of occupational skin diseases, the senior author invited collaborators, Dr. TULIPAN and Dr. PECK for the second edition and Dr. TULIPAN and Dr. BIRMINGHAM for the third one. The present edition also contains special chapters on "Occupational Marks" by Dr. FRANCESCO RONCHESE; on "Skin Hazards from Radiation in Atomic Industry, Medicine and Warfare," by Dr. W. D. NORWOOD, and on "Skin Hazards from Surface Active Agents" by ANTHONY M. SCHWARTZ, Ph. D.

The vast material incorporated in this book is presented in 49 chapters. Every chapter brings a selected bibliography. Since litigations often grow out from occupational hazards, the closing chapter of this work deals with the medico-legal aspects of occupational dermatitis. Since the medical mycologist is mainly, if not exclusively, interested in occupational hazards caused by Fungi, this reviewer will consecrate time and space to those chapters dealing with these hazards.

There are two chapters in this work - Chapter 43, dealing with occupational dermatoses from mycotic infections, and Chapter 46 with occupational diseases of the nails, which need closer scrutiny. From mycological standpoint, these two chapters are simply devastating. Compared with other chapters, one feels that they were culled together from many sources without firsthand knowledge of the authors. Thus, they have stated that, "another problem, perhaps the most baffling that confronts the industrial physician, is raised by the dermatophytids that occur in connection with dermatophytosis, or ringworm infection.'' In the last two decades, these "baffling problems'' without these authors' knowledge, have been solved. Further on, one can read, "the optimum medium for monilia is the seborrheic type of skin, subjected to warmth, moisture and maceration.'' From "yeast-infections'' it is stated that, "the cutaneous affections caused by yeast-like organisms are almost as many and varied as those caused by the dermatophytes or ringworm.'' North American blastomycosis "was first studied in 1893 by CAROZZI'' yet, it is notorious that GILCHRIST studied it first and described its causative organism in 1894. Its causative organism is classified as a *Cryptococcus* (*gilchristi*) or, an Oidium (*dermatitidis*) and is a yeast-like fungus.'' The accepted term for this fungus is, "*Blastomyces dermatitidis*'' and is a hyphomycete and not a yeast-like organism, since the classification is based on the saprophytic, cultural form of the parasite. The causative organism of chromoblastomycosis "does not grow by budding in tissue and has a dark, brown color in culture.'' From Coccidioidal granuloma, it is stated that, "JORDAN and WEIDMAN believe that the causative organism of the disease in South America is *Paracoccidioides brasiliensis*.'' This latter organism is the causative agent of Paracoccidioidal granuloma which is an entirely different condition. From "nocardiosis cutis", it is stated that "clinically, the disease is identical with sporotrichosis, but is caused by a different fungus.'' "Blastomycosis occurs in persons whose work brings them in contact with animals,'' can be found in another paragraph. About sugar cane handlers, we are informed that "SERRA has demonstrated that the eruption is due to the spores of a toadstool (sic!), which develop in the cane stalks when piled up in humid places. He has classified this toadstool in the family of Ustilaginae, under the name of *Ustilago hypodites*, which some authors consider as *Sporotrichum dermatodes* (sic!).'' From *Scopulariopsis brevicaulis* we learn that it "may infect the nails or produce a vesicular eruption of the skin; deep lesions such as nodules, granulomata or an ulcerative process'' . . . . a statement which has no foundation whatsoever. Needless to say, the above quotations could have been continued for quite a length, however, these examples shall suffice. Contrary to Basel Nomenclature of Anatomy, we read about "internatal cleft'' which one could not fathom at first sight in that it is the intergluteal cleft. One reads in further paragraphs about "onyxitis'', an archaic term, which in present terminology is onychia. Specific fungus terms are often misspelled: "Phialophera'' instead of *Phialophora*, "Cereosporella'' instead of "*Cercosporella*.''

"Peasants working in the beet fields in the neighborhood of Prague, have been affected with blastomycosis of the hands and feet, and true Blastomycetes have been cultivated from the surface of the growing beet plant.'' This statement should prove the etiologic connections between skin affection and the beet plants! Onychia is attributed "to a parasite which is considered a spore form of a toadstool belonging to the Mucoraceae.''

Despite the reassurance that the third edition was "thoroughly revised", the errors and misstatements in 1957, are exactly the same as in 1947. On page 723, (3rd Ed., 1957) we read: "A white powder from the canes or, rhizomes of large reeds of 'Povrence' (*Arundo donax*)" .... On page 649, (2nd Ed., 1947) one reads; "A white powder from the canes or, rhizomes of large reeds of 'Provence' (*Arundo donax*) ...... "Povrence" and, "Provence" should read "POACEA," a family of grasses ...

Concerning the occupational involvement of the nails, the authors have very hazy ideas based upon a succession of notorious errors culled from the literature twenty years ago. It would be completely useless to go into further discussion on this topic.

The height of carelessness and superficiality is reached by the caption of an illustration. One and the same illustration is used in the 2nd and 3rd editions. The legend in the 2nd edition, page 634, reads: "Sporotrichosis in a Brazilean farmer"; the legend in the 3rd edition, page 705, reads, "South American blastomycosis in a Brazilean farmer". Should the reader decide which caption is right?! It is too painful to continue with these quotations ...

Even though these two chapters thoroughly reviewed herein make only a small portion of the whole work, their importance far outweighs their volume. From the medico-legal standpoint, alleged fungus diseases of the claimants are with great frequency, the centerpiece of litigation.

In Chapter 48, "Medico-legal Aspects of Occupational Dermatitis", there is no clear cut differentiation between allergic type of contact dermatitis, nummular eczema, and pompholyx of the hands, the most frequent sources of controversy coming before the Industrial Commission.

The selection of the bibliography in the chapters discussed is poor. The authors have hardly added anything to that of the 3rd edition compared with the 2nd. They have overlooked (if they looked for them at all) the most important contributions concerning the afflictions of the hands in relation to occupational hazards, being their most frequent localization.

It is surprising that the chapters discussed herein have appeared in the third edition with all of the errors, misprints and misstatements without "thorough revision". These chapters need not alone a revision, but a thorough rewriting from the hand of a man who is an expert not only in medical mycology, but in dermatology in general and in occupational hazards of the skin in special. In their present form and with their present contents, they are wasted efforts of the authors and completely useless. The production of the book as other publications of Lea and Febiger, shows a high standard.

TIBOR BENEDEK

C. S. Holton, G. W. Fischer, R. W. Fulton, Helen Hart and S. E. A. McCallan, Editors: *Plant Pathology, Problems and Progress* 1908—1958. The University of Wisconsin Press for The American Phytopathological Society, Madison, 1959. p.p. xix, 588, 39 figures, 32 plates, 52 tables. $8.50.

In 1908 the American Phytopathological Society was founded in Baltimore. The fiftieth anniversary celebration was held in conjunction with the annual meeting of the American Institute of Biological Sciences at Bloomington, Indiana. The program of the Society included a session for contributed papers, but was primarily devoted to a series of seven major addresses on historical and developmental aspects of plant pathology and nine symposia on major fields in plant pathology. The 51 papers presented in these ten categories were brought together in this anniversary volume.

It would add little to this review to catalogue the titles of the 51 chapters or to list the 75 authors, discussants, introducers, or authors of symposium epilogues. No more distinguished list of writers has presented summaries of their life work in a single book covering most facets of a single field in biology in recent times. No effort was spared in bringing together workers from all over the world for a week of exchanging ideas, and meeting those whose names had only been signatures on letters or words at the head of an article on a printed page.

The first seven chapters bring together major lectures presented at incidental times throughout the week. Basically the group of lectures brings together the history of plant pathology, the development of host resistance studies, chemical control of plant disease, the international nature of plant disease study and control, the place of plant pathology in the scientific and social development of the world, and the status of plant pathology in biology and agriculture in relation to future outlooks.

In the first planned symposium, the physiology of parasitism is considered from points of view of penetration and infection of the host, toxins and other metabolites, pectin enzymes, host factors, metabolism of obligate parasitism and the application of biochemical principles in the study of parasitism.

Mechanisms governing pathogenicity and disease resistance from the genetic point of view are considered in another symposium. Host-parasite interactions in rust and smut diseases are controlled genetically as are those of the *Phytophthora* late blight and the apple scab. Mechanisms of variation in *Fusarium oxysporum* are summarized. Induced mutations in host plants as well as in pathogens are described.

Fungicides are considered from the points of view of plant chemotherapy, uptake and translocation of organic chemicals by higher plants, their commercial development and evaluation. Chemical structure of fungicides is related to their activity, explained by the use of dithiocarbamic acid derivatives as examples, and by a discussion of physical properties and reactivities of fungicides.

Soil microbiology and root disease fungi are considered from many points of view based on the biology and ecology of such organisms. Their distribution and detection, growth and survival, parasitism and pathogenesis are described. Rhizosphere microorganisms and other soil organisms have specific effects on such fungi.

Nematology is given a prominent place in the book. Here the history, ecological relationships, and variation in nematodes are considered before developing the topics of the mechanism of nematode injury to plants and progress in the chemical control of nematodes.

Virology is represented by two symposia, one on structure of viruses, the other on multiplication of plant viruses. Structure is determined either by electron microscopy or X-ray diffraction. The role of nucleic acid in infection with tobacco mosaic virus is described. Virus infectivity and strain differences are related to chemical composition and structure of the virus. Tobacco mosaic virus is used to demonstrate the biochemistry of the synthesis and biological activity of a virus, while the role and occurrence of noninfectious proteins are considered in relation to virus synthesis. Finally, the establishment and development of infection, and the interaction of viruses in the host are described.

The concluding symposium considers epidemiology. Such problems as spore liberation and dispersal, insect vectors and the spread of viruses in the field, microclimate and infection, plant disease forecasting and problems of systemic infection are described.

Biologists dealing with soil microorganisms, nematodes, viruses, disease resistance and many other fields will find many areas of interest for their own work in this book. It is a general reference work that all people working with living material will find a useful addition to their library.

WM. BRIDGE COOKE

Robert A. Taft Sanitary Engineering Center
Cincinnati, Ohio

Microbiology, Yesterday and Today Edited by VERNON BRYSON Published by Institute of Microbiology, Rutgers University.

This book is a record of the proceedings presented on the occasion of a symposium held at the Institute of Microbiology at Rutgers University in honor of the 70th birthday of Dr. SELMAN A. WAKSMAN. The various sections are presented in the form of a historical review of certain disciplines in biology which have been influenced by microbiological research or have themselves influenced the course of development in microbiology.

J. H. QUASTEL reviews the development of microbial biochemistry, briskly walking through its history with all the dignity and authority that is properly his. He points out the fruitfulness of the use of microorganisms for the study of biochemical pathways and emphasizes the extraordinary versatility of these organisms in adapting or otherwise modifying themselves to be able to act on various substrates, so giving rise to new and interesting biochemical products. Such a versatility is impressive not only with regards to the accomplishments to date but, as QUASTEL points out, is particularly intriguing with regard to what might be accomplished in the future.

H. B. WOODRUFF discusses the potentiality of antibiotics not only in their role as antimicrobial agents, but because of their high degree of specificity in acting on metabolic pathways, may reveal new pathways in microorganisms not readily discernable in the past. In addition, they still offer great potentiality for the therapy of various non-bacterial diseases such as cancer, virus diseases, helminth infestations, and numerous other applications such as plant pathology and food preservation.

HEIDELBERGER'S entertaining paper on episodes in immunochemistry pokes some good-natured fun at the hands which probably originally fed him, namely the disciples of the serological titer. This paper traces the evolution of immunochemistry, citing the experiences of many pioneers in this field, and presenting its future as HEIDELBERGER visualizes it.

Anyone faced with the problem of discussing bacterial classification, as is S. T. COWAN, will find himself surrounded by discension on all sides. This paper is most significant in pointing out the various criteria: physical, habitat, serological, pathogenic, and numerous others which have been and can be used in the classification of microorganisms. COWAN blames the serologists for the confused state of the antigenic criteria for *Salmonella* classification. Should not one more properly blame the *Salmonella*?

The problem of classification as complicated by variations and mutations is also discussed by COWAN, although perhaps not as objectively as one might wish. I assume he might have tongue in cheek when he proposes a return to the simple taxonomy where "most rod shape bacteria are species of one genus". This would probably precipitate a chaos similar to the one he decries which was perpetuated by the immunologists and serologists working with the *Salmonella*. That such a simplified attempt at classification is to be avoided is especially borne out by the paper given by VERNON BRYSON.

BRYSON authoritatively reveals the tremendous latitude of microbial variability and mutation. He would like to change the old dictum that "like begets like" to "like begets similar". He presents good evidence to substantiate such a proposal. His report particularly emphasizes the pioneering studies in microbial genetics by so many workers who paved the way in this discipline with far more understanding than they are given credit for by our contemporaries. Although hindsight is very simple and relationships are easier to make after the fact, it is quite clear that numerous pin points of understanding were beginning to appear in the latter part of the 19th century and early 20th century in studies which are rarely credited by contemporary microbial genticists.

We were particularly intrigued with BRYSON's reference to the work of a little known Mme. HENRI who in 1914 described the mutagenic action of ultraviolet radiation, an observation which became the object of a patent application not too long ago.

GEORGE K. SKRIABIN of the Russian Academy of Sciences reviews the roles of Russian microbiologists such as METCHNIKOV, WINOGRADSKY, OMELIANSKI, NADSON, the very talented KRASSILNIKOV, and others.

The coda by Dr. WAKSMAN himself reviews microbiology yesterday and today. He points out the universality of contributions that were made to these fields in that this interrelationship of work was the necessary feature of success of the whole field itself.

The book will be of interest to all professional microbiologists and workers in allied fields.

GEORGE H. SCHERR

CONSOLIDATED LABORATORIES, INC.
Chicago Heights, Illinois

The Human Integument. Normal and Abnormal. Edited by STEPHEN ROTHMAN. Publication no. 54 of the American Association for the Advancement of Science. 260 pp., Illustrated. Washington, D. C., 1959. Price: $ 6.75.

This Symposium of the Association for the Advancement of Science, cosponsored by the American Medical Association's Committee on Cosmetics and the Society for Investigative Dermatology, represents an endeavor of the Association to maintain close contact with the developments in the basic biological sciences which have an ever increasing impact on the progress of medical sciences.

From the four chapters of the Symposium the medical mycologist is directly interested only in the first chapter which contains the following contributions: Relation of the Anatomy of Normal and Abnormal Skin to its Protective Function, by R. B. STOUGHTON, Protection against the Transfer of Matter through the Skin by R. D. GRIESEMER, and Protection against the Invasion of Bacteria and Fungi, by I. H. BLANK. Each contribution has its own well selected bibliography. The text is well written, clear and concise and most informative. The production of this volume is excellent. It can be well recommended to medical mycologists.

TIBOR BENEDEK

C. M. PALMER, *Algae in Water Supplies*. Public Health Service Publication No. 657, 1959. pp viii, 88. 13 tables, 55 text figures, 6 plates. Supt. of Documents, U. S. Govt. Printing Office. $1.00.

One justification for presenting a work on algae to the readers of this journal is the fact that in some phases of applied mycology one has need to know the algal host of a fungus, a species causing an algal bloom which may be controlled by a fungus, the source of nutrients for a fungal population dependent on algal cells, or one may want to know the types of organisms living as fungal partners in a complex population in fresh water supplies.

An introductory chapter by C. M. TARZWELL explains the importance of algae in water supplies. The twelve additional chapters develop the problems related to the significance of algae in water supplies.

A description of methods of identification, together with line drawings of critical structures, is accompanied by an alphabetical list of important species indicating the morphological group and ecology of each. Old and new names for 38 categories of common algae are tabulated. In the appendix is a dichotomous key using 289 pairs of characters which will help in identification.

Six color plates illustrate 129 species of algae grouped according to the six habitat types. These plates by HAROLD WALTERS accompany chapters on taste and odor algae, filter clogging algae, polluted water algae, clean water algae, plankton and other surface water algae, and algae attached to reservoir walls.

A tabulation of algae causing other problems in water supplies includes discussions of slime producing algae, algae causing coloration of water, corrosion of concrete and steel, presistent in water supplies, interfering with coagulation, causing natural softening of water, and toxic algae. Slime formation, coloration, corrosion and toxicity are emphasized in the accompanying text.

That algae are useful in water supplies is indicated by a tabulation of species useful as water source indicators, indicators of temperature range and acidity, species indicating progress of change in sewage oxidation ponds, and those indicating various types of industrial wastes. Accompanying text material describes industrial uses for algae including potential uses of growth in sewage treatment, and physiological factors making various species useful industrial waste indicators.

Procedures for enumeration of algae in water are given. Each method of enumeration may be applicable to a different use for which the information is required.

Need for and methods of control of algae in raw water supplies, water treatment plants, and distribution systems are considered. Once an interference organism is known control methods may be chemical, mechanical, or related to the construction of the particular installation for which treatment is required.

In addition to the keys and indices, the Appendix includes a glossary of 190 terms and a bibliography of 146 titles.

This is a useful reference for all microbiologists working with fresh water problems.

Robert A. Taft Sanitary Engineering Center
Cincinnati, Ohio

WM. BRIDGE COOKE

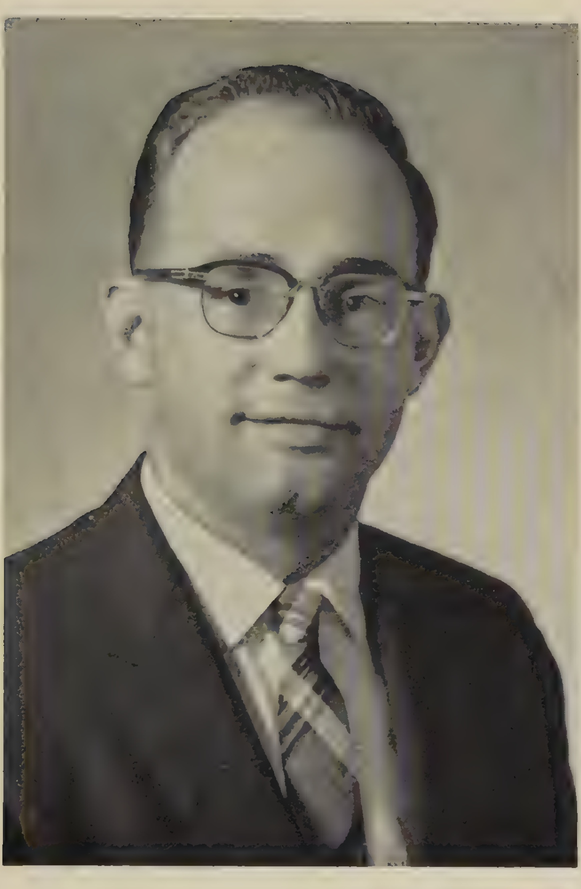
FUNDER, SIGURD: Practical Mycology. Manual for Identification of Fungi, 146 pp. 123 figures in black & white, Hafner Publishing Co., 31 East 10th Street, New York 3, N.Y. 1953. Price $ 6.50.

This book is designed to serve as an introduction to mycology for beginners and as an aid for those who without any special botanical knowledge are interested in identifying the most common fungi they meet with in the course of their work.

The text falls into three parts: 1) Introduction to mycology; 2) The identification of fungi by microscopic examination; 3) Table of classification. The excellent illustrative material derived mainly from J. W. WILSON & O. A. PLUNKETT: "Practical Mycology. The Identification of Fungi by Microscopic Examination". Furthermore, M. A. GORDON'S: "Key to the Human Mycoses" is also included in this volume.

The basic idea of this useful volume is sound. The illustrative material is very excellent, the accompanying text is clear and concise. As an introductory work it will be most useful in general mycology as well as for medical mycologists and plant pathologists. It is hoped that this handy volume won't be missing on the bookshelf of a mycological laboratory of any description. The production of the book is highly satisfactory.

TIBOR BENEDEK



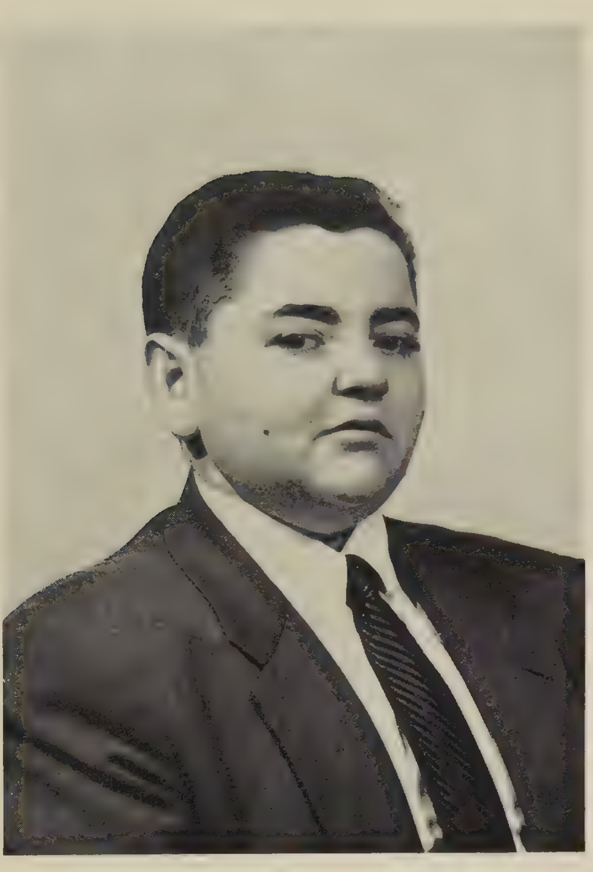
E. S. BENEKE

Dept. of Botany and Plant Pathology

Michigan State University East Lansing, Mich., U.S.A.

Gallery of Contemporary Noted Mycologists

73

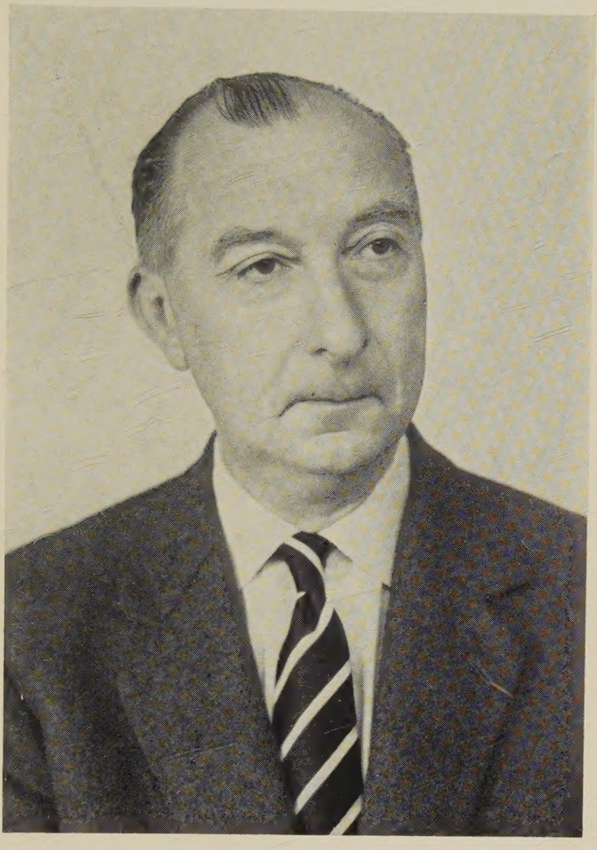


MARVIN A. RICH

Sloan - Kettering Institute for Cancer Research
Division of Biophysics, New York, N.Y., U.S A.

Gallery of Contemporary Noted Mycologists

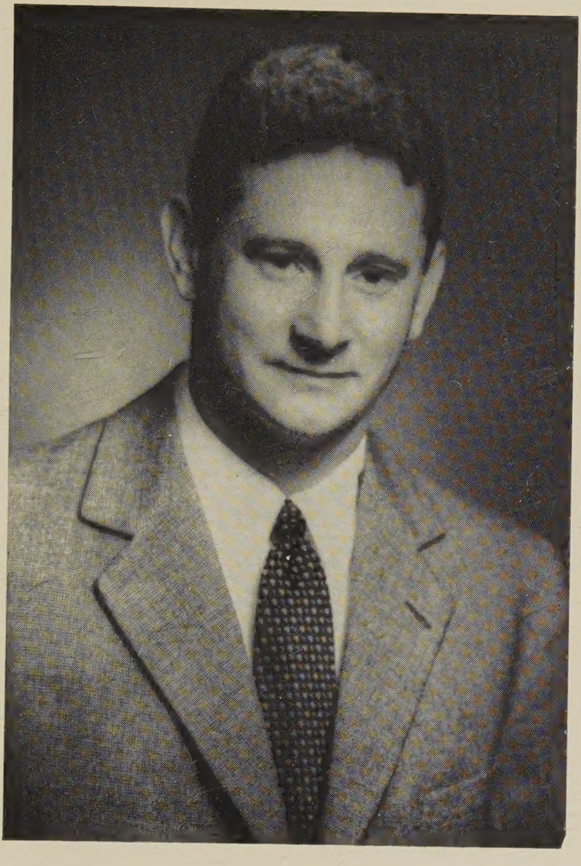
74



R. SARTORY

Institut de Bactériologie et de Cryptogamie
Faculté de Pharmacie, Université de Strasbourg, France

Gallery of Contemporary Noted Mycologists
75



A. M. KLIGMAN

Duhring Laboratories, Dept. of Dermatology
University of Pennsylvania, Philadelphia, Pa., U.S.A.

Gallery of Contemporary Noted Mycologists
76